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FILE 'HOME' ENTERED AT 15:29:08 ON 07 MAY 2004

COST IN U.S. DOLLARS                                    SINCE FILE      TOTAL
                                                           ENTRY    SESSION
FULL ESTIMATED COST                                         0.21       0.21

=> d his

     (FILE 'HOME' ENTERED AT 15:29:08 ON 07 MAY 2004)

=> fil .bec
COST IN U.S. DOLLARS                                    SINCE FILE      TOTAL
                                                           ENTRY    SESSION
FULL ESTIMATED COST                                         0.21       0.21

FILES 'MEDLINE, SCISEARCH, LIFESCI, BIOTECHDS, BIOSIS, EMBASE, HCAPLUS, NTIS,
       ESBIOBASE, BIOTECHNO, WPIDS' ENTERED AT 15:29:44 ON 07 MAY 2004
ALL COPYRIGHTS AND RESTRICTIONS APPLY.  SEE HELP USAGETERMS FOR DETAILS.

11 FILES IN THE FILE LIST

=> act amylase/a
L1   (        37)SEA FILE=MEDLINE ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUTAN
L2   (        55)SEA FILE=SCISEARCH ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUT
L3   (        22)SEA FILE=LIFESCI ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUTAN
L4   (        63)SEA FILE=BIOTECHDS ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUT
L5   (        40)SEA FILE=BIOSIS ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUTANT
L6   (        30)SEA FILE=EMBASE ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUTANT
L7   (        95)SEA FILE=HCAPLUS ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUTAN
L8   (         0)SEA FILE=NTIS ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUTANT#
L9   (        20)SEA FILE=ESBIOBASE ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUT
L10  (        25)SEA FILE=BIOTECHNO ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUT
L11  (        31)SEA FILE=WPIDS ABB=ON  ALPHA AMYLASE# AND BACILLUS AND (MUTANT#
L12  (       418)SEA ALPHA AMYLASE# AND BACILLUS AND (MUTANT# OR VARIANT#) AND (
L13          184 DUP REM L12 (234 DUPLICATES REMOVED)

=> d tot

L13  ANSWER 1 OF 184  EMBASE  COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED.
     on STN
TI   Role of Phe283 in enzymatic reaction of cyclodextrin glycosyltransferase
     from alkalophilic Bacillus sp.1011: Substrate binding and
     arrangement of the catalytic site.
SO   Protein Science, (2004) 13/2 (457-465).
     Refs: 24
     ISSN: 0961-8368  CODEN: PRCIEI
AU   Kanai R.; Haga K.; Akiba T.; Yamane K.; Harata K.
AN   2004045444  EMBASE

L13  ANSWER 2 OF 184  SCISEARCH  COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 1
TI   Improved thermostability of Bacillus circulans
     cyclodextrin glycosyltransferase by the introduction of a salt bridge
SO   PROTEINS-STRUCTURE FUNCTION AND GENETICS, (1 JAN 2004) Vol. 54, No. 1, pp.
     128-134.
     Publisher: WILEY-LISS, DIV JOHN WILEY & SONS INC, 605 THIRD AVE, NEW YORK,
     NY 10158-0012 USA.
     ISSN: 0887-3585.
AU   Leemhuis H; Rozeboom H J; Dijkstra B W; Dijkhuizen L (Reprint)
AN   2004:87846  SCISEARCH

L13  ANSWER 3 OF 184  BIOSIS  COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
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stability or thermostability or specific activity or calcium)

TI Improved **thermostability** of **Bacillus** circulans cyclodextrin glycosyltransferase by the introduction of a salt bridge.
SO Proteins Structure Function and Bioinformatics, (January 1 2004) Vol. 54, No. 1, pp. 128-134. print.
AU Leemhuis, Hans; Rozeboom, Henriette J.; Dijkstra, Bauke W.; Dijkhuizen, Lubbert [Reprint Author]
AN 2004:126635 BIOSIS

L13 ANSWER 4 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Evaluating the efficacy of molecule against target population including toxin-resistant pest strain, by determining susceptible pest strain, selecting resistant strain, and evaluating efficacy of resistant strain with molecules;
expression profiling of selected genes after exposure to toxin
AU PITTENDRIGH B R; MURDOCK L L; GAFFNEY P J
AN 2003-21526 BIOTECHDS
PI WO 2003060463 24 Jul 2003

L13 ANSWER 5 OF 184 MEDLINE on STN DUPLICATE 2
TI Kinetic stabilization of **Bacillus** licheniformis **alpha-amylase** through introduction of hydrophobic residues at the surface.
SO Journal of biological chemistry, (2003 Mar 28) 278 (13) 11546-53. Journal code: 2985121R. ISSN: 0021-9258.
AU Machius Mischa; Declerck Nathalie; Huber Robert; Wiegand Georg
AN 2003150207 MEDLINE

L13 ANSWER 6 OF 184 Elsevier BIOBASE COPYRIGHT 2004 Elsevier Science B.V. on STN
AN 2003153274 ESBIOBASE
TI Conversion of cyclodextrin glycosyltransferase into a starch hydrolase by directed evolution: The role of alanine 230 in acceptor subsite +1
AU Leemhuis H.; Rozeboom H.J.; Wilbrink M.; Euverink G.-J.W.; Dijkstra B.W.; Dijkhuizen L.
CS L. Dijkhuizen, Department of Microbiology, Groningen Biomol. Sci./Biotech. I., University of Groningen, Kerklaan 30, 9751 NN Haren, Netherlands.
E-mail: L.Dijkhuizen@biol.rug.nl
SO Biochemistry, (24 JUN 2003), 42/24 (7518-7526), 45 reference(s) CODEN: BICHAW ISSN: 0006-2960
DT Journal; Article
CY United States
LA English
SL English

L13 ANSWER 7 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI Directed evolution of Thermus maltogenic amylase toward enhanced thermal resistance
SO APPLIED AND ENVIRONMENTAL MICROBIOLOGY, (AUG 2003) Vol. 69, No. 8, pp. 4866-4874.
Publisher: AMER SOC MICROBIOLOGY, 1752 N ST NW, WASHINGTON, DC 20036-2904 USA.
ISSN: 0099-2240.
AU Kim Y W; Choi J H; Kim J W; Park C; Kim J W; Cha H J; Lee S B; Oh B H; Moon T W; Park K H (Reprint)
AN 2003:708604 SCISEARCH

L13 ANSWER 8 OF 184 MEDLINE on STN DUPLICATE 3
TI A thermoacidophilic endoglucanase (CelB) from Alicyclobacillus acidocaldarius displays high sequence similarity to arabinofuranosidases belonging to family 51 of glycoside hydrolases.
SO European journal of biochemistry / FEBS, (2003 Sep) 270 (17) 3593-602. Journal code: 0107600. ISSN: 0014-2956.
AU Eckert Kelvin; Schneider Erwin

AN 2003383578 MEDLINE

L13 ANSWER 9 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 4
TI Improving the **thermostability** of raw-starch-digesting amylase from a Cytophaga sp by site-directed mutagenesis
SO APPLIED AND ENVIRONMENTAL MICROBIOLOGY, (APR 2003) Vol. 69, No. 4, pp. 2383-2385.
Publisher: AMER SOC MICROBIOLOGY, 1752 N ST NW, WASHINGTON, DC 20036-2904 USA.
ISSN: 0099-2240.
AU Shiau R J; Hung H C; Jeang C L (Reprint)
AN 2003:326818 SCISEARCH

L13 ANSWER 10 OF 184 MEDLINE on STN DUPLICATE 5
TI Directed evolution of a bacterial **alpha-amylase**: toward enhanced pH-performance and higher **specific activity.**
SO Protein science : a publication of the Protein Society, (2003 Oct) 12 (10) 2141-9.
Journal code: 9211750. ISSN: 0961-8368.
AU Bessler Cornelius; Schmitt Jutta; Maurer Karl-Heinz; Schmid Rolf D
AN 2003441863 IN-PROCESS

L13 ANSWER 11 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 6
TI Effects of **mutant** thermostable **alpha-amylases** on rheological properties of wheat dough and bread
SO CEREAL CHEMISTRY, (NOV-DEC 2003) Vol. 80, No. 6, pp. 722-727.
Publisher: AMER ASSOC CEREAL CHEMISTS, 3340 PILOT KNOB RD, ST PAUL, MN 55121-2097 USA.
ISSN: 0009-0352.
AU Maeda T; Hashimoto T; Minoda M; Tamagawa S; Morita N (Reprint)
AN 2003:1021679 SCISEARCH

L13 ANSWER 12 OF 184 MEDLINE on STN DUPLICATE 7
TI Identification of essential histidine residues in a recombinant **alpha-amylase** of thermophilic and alkaliphilic **Bacillus** sp. strain TS-23.
SO Extremophiles : life under extreme conditions, (2003 Dec) 7 (6) 505-9.
Journal code: 9706854. ISSN: 1431-0651.
AU Chang Chen-Tien; Lo Huei-Fen; Chi Meng-Chun; Yao Chia-Yu; Hsu Wen-Hwei; Lin Long-Liu
AN 2003584416 IN-PROCESS

L13 ANSWER 13 OF 184 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN DUPLICATE 8
TI α **-amylase** from **Bacillus** licheniformis **mutants** near to the catalytic site: Effects on hydrolytic and transglycosylation activity.
SO Protein Engineering, (1 Jul 2003) 16/7 (505-514).
Refs: 61
ISSN: 0269-2139 CODEN: PRENE
AU Rivera M.H.; Lopez-Munguia A.; Soberon X.; Saab-Rincon G.
AN 2003349435 EMBASE

L13 ANSWER 14 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 9
TI **alpha-amylases** of medical and industrial importance
SO JOURNAL OF MOLECULAR STRUCTURE-THEOCHEM, (29 DEC 2003) Vol. 666, pp. 487-498.
Publisher: ELSEVIER SCIENCE BV, PO BOX 211, 1000 AE AMSTERDAM, NETHERLANDS.
ISSN: 0166-1280.
AU Kandra L (Reprint)
AN 2004:197559 SCISEARCH

L13 ANSWER 15 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Oxidative stabilization of an alkaliphilic **Bacillus** . **alpha.-amylase** by replacing a single specific methionine residue by site-directed mutagenesis
SO Journal of Applied Glycoscience (2003), 50(3), 367-372
CODEN: JAGLFX; ISSN: 1344-7882
AU Hagihara, Hiroshi; Hatada, Yuji; Ozawa, Tadahiro; Igarashi, Kazuaki; Araki, Hiroyuki; Ozaki, Katsuya; Kobayashi, Tohru; Kawai, Shuji; Ito, Susumu
AN 2003:685189 HCAPLUS
DN 139:257261

L13 ANSWER 16 OF 184 MEDLINE on STN DUPLICATE 10
TI Hyperthermostabilization of **Bacillus** licheniformis **alpha** -**amylase** and modulation of its **stability** over a 50 degrees C temperature range.
SO Protein engineering, (2003 Apr) 16 (4) 287-93.
Journal code: 8801484. ISSN: 0269-2139.
AU Declerck Nathalie; Machius Mischa; Joyet Philippe; Wiegand Georg; Huber Robert; Gaillardin Claude
AN 2003215498 IN-PROCESS

L13 ANSWER 17 OF 184 MEDLINE on STN DUPLICATE 11
TI Replacement of methionine 208 in a truncated **Bacillus** sp. TS-23 **alpha-amylase** with oxidation-resistant leucine enhances its resistance to hydrogen peroxide.
SO Current microbiology, (2003 Mar) 46 (3) 211-6.
Journal code: 7808448. ISSN: 0343-8651.
AU Lin Long-Liu; Lo Huei-Fen; Chiang Wen-Ying; Hu Hui-Yu; Hsu Wen-Hwei; Chang Chen-Tien
AN 2003055934 MEDLINE

L13 ANSWER 18 OF 184 MEDLINE on STN
TI Engineering cyclodextrin glycosyltransferase into a starch hydrolase with a high exo-specificity.
SO Journal of biotechnology, (2003 Aug 15) 103 (3) 203-12.
Journal code: 8411927. ISSN: 0168-1656.
AU Leemhuis Hans; Kragh Karsten M; Dijkstra Bauke W; Dijkhuizen Lubbert
AN 2003358258 MEDLINE

L13 ANSWER 19 OF 184 MEDLINE on STN
TI Three-dimensional structure and substrate binding of **Bacillus** stearothermophilus neopullulanase.
SO Journal of molecular biology, (2003 Feb 7) 326 (1) 177-88.
Journal code: 2985088R. ISSN: 0022-2836.
AU Hondoh Hironori; Kuriki Takashi; Matsuura Yoshiki
AN 2003040244 MEDLINE

L13 ANSWER 20 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI Protein engineering of detergent **alpha-amylases**
SO TRENDS IN GLYCOSCIENCE AND GLYCOTECHNOLOGY, (MAR 2003) Vol. 15, No. 82, pp. 101-114.
Publisher: FCCA-FORUM CARBOHYDRATES COMING AGE, C/O GAKUSHIN PUBLISHING CO LTD 1-1-8 TARUMI-CHO, SUITA 564-0062, OSAKA, JAPAN.
ISSN: 0915-7352.
AU Igarashi K (Reprint); Hagihara H; Ito S
AN 2003:396985 SCISEARCH

L13 ANSWER 21 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Novel **variant** of parent Termamyl-like **alpha-amylase** useful for starch liquefaction, washing and/or dishwashing, has **alpha-amylase** activity and exhibits altered properties relative to the parent **alpha-amylase** ;

vector-mediated gene transfer and expression in host cell for recombinant protein production
AU SVENDSEN A; ANDERSEN C; THISTED T; VON DER OSTEN C
AN 2003-09677 BIOTECHDS
PI WO 2002092797 21 Nov 2002

L13 ANSWER 22 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI KSM-K36 or KSM-K38 **variant** from **Bacillus** for cleaning dishes, textile desizing, starch liquefaction and ethanol production has **alpha-amylase** activity;
plasmid-mediated recombinant **mutant** enzyme gene transfer and expression in **Bacillus** sp.
AU ANDERSEN C
AN 2002-16321 BIOTECHDS
PI WO 2002031124 18 Apr 2002

L13 ANSWER 23 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI **Variant** of parent Termamyl-like **alpha amylase** , useful in detergent compositions, for starch liquefaction, ethanol production, washing and/or dish washing, and textile desizing;
recombinant enzyme production, vector expression in host cell, polymerase chain reaction and mutagenesis
AU THISTED T; KJAERULFF S; ANDERSEN C; FUGLSANG C C
AN 2002-12006 BIOTECHDS
PI WO 2002010355 7 Feb 2002

L13 ANSWER 24 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Novel **variant** of cell-wall degrading enzyme having beta-helix structure, specifically **variant** of wild-type pectate lyase useful in textile, detergent and cellulose fiber processing and in wine and juice processing;
plasmid-pMB54-mediated recombinant pectate-lyase, **alpha-amylase**, chloramphenicol-acetyltransferase fusion protein gene transfer and expression in **Bacillus** subtilis and transgenic plant for use as a feed-addictive and in thepaper industry
AU SCHUELEIN M; GLAD S O S; ANDERSEN C; FRANDSEN T P
AN 2002-12004 BIOTECHDS
PI WO 2002006442 24 Jan 2002

L13 ANSWER 25 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI New **mutant alpha-amylase**, useful in detergent compositions, comprises increased productivity when prepared recombinantly and better resistance to heat;
recombinant enzyme protein production via plasmid expression in bacterium cell, for surfactant composition and starch liquefaction
AU ARAKI H; HAGIHARI H; HAYASHI Y; ENDO K; IGARASHI K; OZAKI K
AN 2002-15685 BIOTECHDS
PI EP 1199356 24 Apr 2002

L13 ANSWER 26 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI α -**Amylases** and α -**amylase** **variants** with improved properties for commercial uses
SO U.S., 64 pp., Cont.-in-part of U.S. 6,187,576.
CODEN: USXXAM
IN Svendsen, Allan; Borchert, Torben Vedel; Bisgard-Frantzen, Henrik; Outtrup, Helle; Nielsen, Bjarne Ronfeldt; Nielsen, Vibeke Skovgaard; Hedegaard, Lisbeth
AN 2002:236435 HCAPLUS
DN 136:259230

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI US 6361989	B1	20020326	US 1999-290734	19990413
US 6187576	B1	20010213	US 1998-170670	19981013
WO 2000060060	A2	20001012	WO 2000-DK149	20000328

WO 2000060060 A3 20010419
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

BR 2000009392	A	20020108	BR 2000-9392	20000328
EP 1173554	A2	20020123	EP 2000-912416	20000328
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO				
JP 2002540786	T2	20021203	JP 2000-609552	20000328
US 6528298	B1	20030304	US 2000-545586	20000407
US 2003211958	A1	20031113	US 2002-327837	20021223

L13 ANSWER 27 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Recombinant **mutant** alkalophilic **Bacillus** . **alpha.-amylase** with improved **thermostability**, recombinant expression, and detergent use
SO Jpn. Kokai Tokkyo Koho, 28 pp.
CODEN: JKXXAF
IN Araki, Hiroyuki; Endo, Keiji; Hagiwara, Hiroshi; Igarashi, Kazuaki; Hayashi, Yasuhiro; Ozaki, Katsuya
AN 2002:284478 HCAPLUS
DN 136:305146

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI JP 2002112792	A2	20020416	JP 2000-310605	20001011
US 2002123124	A1	20020905	US 2001-971611	20011009
EP 1199356	A2	20020424	EP 2001-123378	20011010
EP 1199356	A3	20020515		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR				
CN 1348000	A	20020508	CN 2001-141253	20011011

L13 ANSWER 28 OF 184 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI Aqueous liquid or gel type detergent, useful as automatic dishwashing composition, comprises boric acid or born compound, polyhydroxy compound, **calcium** ions and **alpha-amylase** enzyme,.
PI WO 2002068575 A1 20020906 (200305)* EN 36 C11D003-386
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TR TZ UG ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CO CR CU CZ DE DK DM DZ EC EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ PH PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG UZ VN YU ZA ZW

US 2002183226	A1	20021205 (200305)		C11D003-386
EP 1373452	A1	20040102 (200409)	EN	C11D003-386
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI TR				

IN KASTURI, C; SONG, B X; WANDSTRAT, M E; WANDSRAT, M E

L13 ANSWER 29 OF 184 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI Detergent composition for removing starch-containing stains on fabrics, comprises cyclodextrin glucanotransferase enzyme and detergent ingredient which is non-ionic surfactant, protease and bleaching agent.
PI WO 2002002725 A1 20020110 (200227)* EN 97 C11D003-386
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TR TZ UG ZW
W: AE AL AM AT AU AZ BA BB BG BR BY CA CH CN CR CU CZ DE DK DM EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS

LT LU LV MA MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL
TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW

US 2002032142	A1	20020314	(200227)			C12S009-00
AU 2000060630	A	20020114	(200237)			C11D003-386
EP 1294844	A1	20030326	(200323)	EN		C11D003-386
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI						
BR 2000017277	A	20030506	(200334)			C11D003-386
CZ 2002004166	A3	20030514	(200337)			C11D003-386
KR 2003010758	A	20030205	(200338)			C11D003-386
HU 2003000967	A2	20030728	(200379)			C11D003-386
JP 2004502831	W	20040129	(200413)		167	C11D003-386

IN PINTENS, A; SMETS, J

L13 ANSWER 30 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI A novel, high performance enzyme for starch liquefaction - Discovery and optimization of a low pH, thermostable **alpha-amylase**
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (19 JUL 2002) Vol. 277, No. 29, pp. 26501-26507.
Publisher: AMER SOC BIOCHEMISTRY MOLECULAR BIOLOGY INC, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814-3996 USA.
ISSN: 0021-9258.
AU Richardson T H (Reprint); Tan X Q; Frey G; Callen W; Cabell M; Lam D; Macomber J; Short J M; Robertson D E; Miller C
AN 2002:614062 SCISEARCH

L13 ANSWER 31 OF 184 MEDLINE on STN DUPLICATE 17
TI Pyrococcus furiosus **alpha-amylase** is stabilized by **calcium** and zinc.
SO Biochemistry, (2002 May 14) 41 (19) 6193-201.
Journal code: 0370623. ISSN: 0006-2960.
AU Savchenko Alexei; Vieille Claire; Kang Suil; Zeikus J Gregory
AN 2002254970 MEDLINE

L13 ANSWER 32 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Selection of a Potent **Bacillus** licheniformis Strain Producing Thermostable Amylase
SO Applied Biochemistry and Microbiology (Translation of Prikladnaya Biokhimiya i Mikrobiologiya) (2002), 38(5), 427-432
CODEN: APBMAC; ISSN: 0003-6838
AU Tsurikova, N. V.; Nefedova, L. I.; Kostyleva, E. V.; Zvenigorodskii, V. I.; Yasinovskii, V. G.; Voeikova, T. A.; Sinitsyn, A. P.
AN 2002:663727 HCAPLUS
DN 137:383856

L13 ANSWER 33 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI Simultaneous inactivation of the wprA and dltB genes of **Bacillus** subtilis reduces the yield of **alpha-amylase**
SO LETTERS IN APPLIED MICROBIOLOGY, (MAY 2002) Vol. 34, No. 6, pp. 394-397.
Publisher: BLACKWELL PUBLISHING LTD, P O BOX 88, OSNEY MEAD, OXFORD OX2 0NE, OXON, ENGLAND.
ISSN: 0266-8254.
AU Stephenson K; Jensen C L; Jorgensen S T; Harwood C R (Reprint)
AN 2002:444881 SCISEARCH

L13 ANSWER 34 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Improvement of **thermostability** of a **calcium**-free . **alpha.-amylase** from an alkaliphilic **Bacillus** sp. by protein engineering
SO Journal of Applied Glycoscience (2002), 49(3), 281-289
CODEN: JAGLFX; ISSN: 1344-7882
AU Hagihara, Hiroshi; Igarashi, Kazuaki; Hayashi, Yasuhiro; Kitayama, Kaori; Endo, Keiji; Ozawa, Tadahiro; Ozaki, Katsuya; Kawai, Shuji; Ito, Susumu
AN 2002:660551 HCAPLUS

DN 137:212837

L13 ANSWER 35 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Protein-engineered **Bacillus** α -**amylases** that have acquired both enhanced **thermostability** and chelator resistance
SO Journal of Applied Glycoscience (2002), 49(3), 257-264
CODEN: JAGLFX; ISSN: 1344-7882
AU Ito, Susumu; Hatada, Yuji; Ozawa, Tadahiro; Hagihara, Hiroshi; Araki, Hiroyuki; Tsujino, Yukiharu; Kitayama, Kaori; Igarashi, Kazuaki; Kageyama, Yasushi; Kobayashi, Tohru; Ozaki, Katsuya
AN 2002:660548 HCAPLUS
DN 137:212836

L13 ANSWER 36 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 18
TI Engineering the **thermostability** of **Bacillus** licheniformis **alpha-amylase**
SO BIOLOGIA, (JAN 2002) Vol. 57, Supp. [11], pp. 203-211.
Publisher: SLOVAK ACADEMIC PRESS LTD, PO BOX 57 NAM SLOBODY 6, 810 05 BRATISLAVA, SLOVAKIA.
ISSN: 0006-3088.
AU Declerck N (Reprint); Machius M; Joyet P; Wiegand G; Huber R; Gaillardin C
AN 2003:91003 SCISEARCH

L13 ANSWER 37 OF 184 MEDLINE on STN DUPLICATE 19
TI Deletion analysis of the C-terminal region of the **alpha-amylase** of **Bacillus** sp. strain TS-23.
SO Archives of microbiology, (2002 Aug) 178 (2) 115-23.
Journal code: 0410427. ISSN: 0302-8933.
AU Lo Huei-Fen; Lin Long-Liu; Chiang Wen-Ying; Chie Meng-Chun; Hsu Wen-Hwei; Chang Chen-Tien
AN 2002369647 MEDLINE

L13 ANSWER 38 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Novel **variant** of parent termamyl-like **alpha-amylase** useful as a component in washing and dishwashing compositions, for textile desizing, for starch liquefaction, and for producing sweeteners and ethanol from starch;
vector plasmid pJE1-mediated recombinant enzyme gene transfer and expression in Escherichia coli, surfactant and polymerase chain reaction for use in starch liquefaction, textile industry, sweetener and ethanolpreparation
AU ANDERSEN C; BORCHERT T V; NIELSEN B R
AN 2002-11532 BIOTECHDS
PI WO 2001066712 13 Sep 2001

L13 ANSWER 39 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI A gene encoding a **mutant alpha-amylase** obtained by making replacement or deletions of amino acid residues in a characteristic sequence;
involving recombinant vector plasmid pHSP-K38, plasmid pHSP-LAMY-mediated gene transfer for expression in host cell
AU Endo K; Igarashi K; Hayashi Y; Hagihara H; Ozaki K
AN 2001-05257 BIOTECHDS
PI EP 1065277 3 Jan 2001

L13 ANSWER 40 OF 184 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN DUPLICATE 22
TI **Mutant alpha-amylase.**
SO Official Gazette of the United States Patent and Trademark Office Patents, (Apr. 3, 2001) Vol. 1245, No. 1. e-file.
CODEN: OGUPE7. ISSN: 0098-1133.
AU Caldwell, Robert M. [Inventor]; Mitchinson, Colin [Inventor]; Ropp, Traci

H. [Inventor, Reprint author]
AN 2001:440007 BIOSIS

L13 ANSWER 41 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 23
TI Production of oxidatively stable **Bacillus** α - **amylase** recombinant **mutants** and their use in detergents and starch liquefaction compositions
SO U.S., 57 pp., Cont.-in-part of U.S. Ser. No. 16,395, abandoned.
CODEN: USXXAM
IN Barnett, Christopher C.; Mitchinson, Colin; Power, Scott D.; Requadt, Carol A.
AN 2001:719022 HCAPLUS
DN 135:285005

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6297037	B1	20011002	US 1994-194664	19940210
CN 1118172	A	19960306	CN 1994-191138	19940210
CN 1104499	B	20030402		
HU 72920	A2	19960628	HU 1995-2364	19940210
HU 219675	B	20010628		
EP 867504	A1	19980930	EP 1998-109967	19940210
EP 867504	B1	20030502		
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE				
AT 175235	E	19990115	AT 1994-909609	19940210
ES 2126743	T3	19990401	ES 1994-909609	19940210
AT 239075	E	20030515	AT 1998-109967	19940210
ES 2198617	T3	20040201	ES 1998-109967	19940210
CZ 293163	B6	20040218	CZ 1995-2057	19940210
US 5824532	A	19981020	US 1995-468220	19950606
US 5849549	A	19981215	US 1995-468698	19950606

L13 ANSWER 42 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Cloning, characterization and use of alkaline α - **amylase** from **Bacillus**
SO PCT Int. Appl., 107 pp.
CODEN: PIXXD2
IN Andersen, Carsten; Outtrup, Helle; Nielsen, Bjarne Roenfeldt; Hoeck, Lisbeth Hedegaard
AN 2001:661564 HCAPLUS
DN 135:223447

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001064852	A1	20010907	WO 2001-DK133	20010228
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				

L13 ANSWER 43 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Method for obtaining proteins having improved **stability** characteristics
SO PCT Int. Appl., 42 pp.
CODEN: PIXXD2
IN Day, Anthony G.; Mitchinson, Colin; Shaw, Andrew
AN 2001:489425 HCAPLUS
DN 135:103326

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001047956	A2	20010705	WO 2000-US33878	20001214

WO 2001047956 A3 20020214
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
EP 1240524 A2 20020918 EP 2000-984363 20001214
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR
JP 2004500815 T2 20040115 JP 2001-549426 20001214

L13 ANSWER 44 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI α -**Amylase variants** with improved detergent performance
SO U.S., 36 pp.
CODEN: USXXAM
IN Svendsen, Allan; Kjaerulff, Soeren; Bisgaard-Frantzen, Henrik; Andersen, Carsten
AN 2001:161441 HCAPLUS
DN 134:190018

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6197565	B1	20010306	US 1998-193068	19981116

L13 ANSWER 45 OF 184 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI New **variant** of Fungamyl-like **alpha-amylase**, useful for production of maltose syrups, includes mutations that improve **stability** against heat and acidic pH.
PI WO 2001034784 A1 20010517 (200138)* EN 47 C12N009-30
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW MZ NL OA PT SD SE SL SZ TR TZ UG ZW
W: AE AG AL AM AT AU AZ BA BB BG BR BY BZ CA CH CN CR CU CZ DE DK DM DZ EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX MZ NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG UZ VN YU ZA ZW
AU 2001012696 A 20010606 (200152) C12N009-30
EP 1230351 A1 20020814 (200261) EN C12N009-30
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI TR
JP 2003513666 W 20030415 (200328) 54 C12N015-09
CN 1390252 A 20030108 (200334) C12N009-30
IN BISGARD-FRANTZEN, H; PEDERSEN, S; SVENDSEN, A

L13 ANSWER 46 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 24
TI Transcripts of the genes sacB, amyE, sacC and csn expressed in **Bacillus** subtilis under the control of the 5 ' untranslated sacR region display different stabilities that can be modulated
SO MICROBIOLOGY-SGM, (MAY 2001) Vol. 147, Part 5, pp. 1331-1341.
Publisher: SOC GENERAL MICROBIOLOGY, MARLBOROUGH HOUSE, BASINGSTOKE RD, SPENCERS WOODS, READING RG7 1AE, BERKS, ENGLAND.
ISSN: 1350-0872.
AU Pereira Y; Chambert R; Leloup L; Daguer J P; Petit-Glatron M F (Reprint)
AN 2001:395189 SCISEARCH

L13 ANSWER 47 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI The deletion of amino-terminal domain in Thermoactinomyces vulgaris R-47 **alpha-amylases**: Effects of domain N on activity, specificity, **stability** and dimerization
SO BIOSCIENCE BIOTECHNOLOGY AND BIOCHEMISTRY, (FEB 2001) Vol. 65, No. 2, pp.

401-408.
Publisher: JAPAN SOC BIOSCI BIOTECHN AGROCHEM, JAPAN ACAD SOC CTR BLDG, 2-4-6 YAYOI BUNKYO-KU, TOKYO, 113, JAPAN.
ISSN: 0916-8451.
AU Yokota T; Tonozuka T; Kamitori S; Sakano Y (Reprint)
AN 2001:218059 SCISEARCH

L13 ANSWER 48 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI Extensive N-glycosylation reduces the thermal **stability** of a recombinant alkalophilic **Bacillus alpha-amylase** produced in Pichia pastoris
SO PROTEIN EXPRESSION AND PURIFICATION, (FEB 2001) Vol. 21, No. 1, pp. 13-23.
Publisher: ACADEMIC PRESS INC, 525 B ST, STE 1900, SAN DIEGO, CA 92101-4495 USA.
ISSN: 1046-5928.
AU Tull D; Gottschalk T E; Svendsen I; Kramhoft B; Phillipson B A; Bisgard-Frantzen H; Olsen O; Svensson B (Reprint)
AN 2001:170633 SCISEARCH

L13 ANSWER 49 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 25
TI α -**Amylase variants** with improved specificity and/or **specific activity**
SO PCT Int. Appl., 78 pp.
CODEN: PIXXD2
IN Andersen, Carsten; Jorgensen, Christel Thea; Bisgard-Frantzen, Henrik; Svendsen, Allan; Kjaerulff, Soren
AN 2000:725751 HCAPLUS
DN 133:292888

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2000060059	A2	20001012	WO 2000-DK148	20000328
WO 2000060059	A3	20010510		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
BR 2000009362	A	20011226	BR 2000-9362	20000328
EP 1165762	A2	20020102	EP 2000-912415	20000328
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO				
JP 2002540785	T2	20021203	JP 2000-609551	20000328
US 6410295	B1	20020625	US 2000-537168	20000329
US 2003044954	A1	20030306	US 2002-146327	20020515

L13 ANSWER 50 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 26
TI **Bacillus** Termamyl-like α -**amylase variants** with improved pH and temperature **stability**
SO PCT Int. Appl., 80 pp.
CODEN: PIXXD2
IN Svendsen, Allan; Kjaerulff, Soren; Bisgard-Frantzen, Henrik; Andersen, Carsten
AN 2000:351648 HCAPLUS
DN 133:14086

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2000029560	A1	20000525	WO 1999-DK628	19991116
W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA,				

MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI,
SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ,
BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE,
DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF,
CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

EP 1131418	A1	20010912	EP 1999-972255	19991116
JP 2002530072	T2	20020917	JP 2000-582544	19991116

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
IE, SI, LT, LV, FI, RO

L13 ANSWER 51 OF 184 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
TI **Mutant alpha-amylase** comprising modification
at residues corresponding to A210, H405 and/or T412 in **Bacillus**
licheniformis.
SO Official Gazette of the United States Patent and Trademark Office Patents,
(June 27, 2000) Vol. 1235, No. 4. e-file.
CODEN: OGUPE7. ISSN: 0098-1133.
AU Day, Anthony G. [Inventor]; Swanson, Barbara A. [Inventor, Reprint author]
AN 2001:113980 BIOSIS

L13 ANSWER 52 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Polypeptides having alkaline α -**amylase** activity
and nucleic acids encoding same
SO PCT Int. Appl., 116 pp.
CODEN: PIXXD2
IN Outtrup, Helle; Hoeck, Lisbeth Hedegaard; Nielsen, Bjarne Ronfeldt;
Borchert, Torben Vedel; Nielsen, Vibeke Skovgaard; Bisgard-Frantzen,
Henrik; Svendsen, Allan; Andersen, Carsten
AN 2000:725752 HCAPLUS
DN 133:292889

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2000060060	A2	20001012	WO 2000-DK149	20000328
WO 2000060060	A3	20010419		

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR,
CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU,
ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU,
LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE,
SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW,
AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE,
DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF,
CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

US 6361989	B1	20020326	US 1999-290734	19990413
BR 2000009392	A	20020108	BR 2000-9392	20000328
EP 1173554	A2	20020123	EP 2000-912416	20000328

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
IE, SI, LT, LV, FI, RO

JP 2002540786	T2	20021203	JP 2000-609552	20000328

L13 ANSWER 53 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Polypeptides having alkaline α -**amylase** activity
and nucleic acids encoding same
SO PCT Int. Appl., 112 pp.
CODEN: PIXXD2
IN Outtrup, Helle; Hoeck, Lisbeth Hedegaard; Nielsen, Bjarne Ronfeldt;
Borchert, Torben Vedel; Nielsen, Vibeke Skovgaard; Bisgard-frantzen,
Henrik; Svendsen, Allan; Andersen, Carsten
AN 2000:725750 HCAPLUS
DN 133:307124

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 2000060058	A2	20001012	WO 2000-DK147	20000328

WO 2000060058 A3 20010412
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
EP 1169434 A2 20020109 EP 2000-912414 20000328
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO
JP 2002540784 T2 20021203 JP 2000-609550 20000328

L13 ANSWER 54 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Maltogenic α -**amylase** three-dimensional structure and its use for the construction of **variants** with improved properties
SO U.S., 97 pp., Cont.-in-part of U.S. Ser. No. 77,795.
CODEN: USXXAM
IN Cherry, Joel; Vendsen, Allan; Andersen, Carsten; Beier, Lars; Frandsen, Torben Peter
AN 2000:891507 HCAPLUS
DN 134:53137

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 6162628	A	20001219	US 1999-386607	19990831
WO 9943794	A1	19990902	WO 1999-DK88	19990226

W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

L13 ANSWER 55 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Recombinant **mutant** alkalophilic **Bacillus** . **alpha.-amylase** with improved **thermostability**, recombinant expression, and detergent use
SO Jpn. Kokai Tokkyo Koho, 12 pp.
CODEN: JKXXAF
IN Igarashi, Kazuaki; Endo, Keiji; Hayashi, Yasuhiro; Hagiwara, Hiroshi; Ozaki, Katsuya
AN 2000:630815 HCAPLUS
DN 133:218513

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 2000245466	A2	20000912	JP 1999-48213	19990225

L13 ANSWER 56 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Novel α -**amylase mutants** of **Bacillus** amyloliquefaciens with appropriate **thermostability** and their use for bakery products
SO Jpn. Kokai Tokkyo Koho, 22 pp.
CODEN: JKXXAF
IN Tamakawa, Shinichiro; Yoshida, Masaharu; Minoda, Masashi; Takahashi, Satoko; Hidaki, Yumiko; Tani, Masakazu; Hashimoto, Tetsushi
AN 2000:316787 HCAPLUS
DN 132:344864

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE

PI	JP 2000135093	A2	20000516	JP 1999-234813	19990820

L13 ANSWER 57 OF 184 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI **Variant** bacterial pullulanases and isoamylases having, e.g. increased **thermostability**, used for converting starch from potatoes into high fructose syrup.
PI WO 2000001796 A2 20000113 (200014)* EN 116 C12N000-00
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL OA PT SD SE SL SZ UG ZW
W: AE AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT UA UG UZ VN YU ZA ZW
AU 9948971 A 20000124 (200027) C12N000-00
EP 1092014 A2 20010418 (200123) EN C12N009-44
R: AL AT BE CH CY DE DK ES FI FR GB GR IE IT LI LT LU LV MC MK NL PT RO SE SI
US 6265197 B1 20010724 (200146) C12N009-44
CN 1309701 A 20010822 (200175) C12N009-44
KR 2001081985 A 20010829 (200215) C12N009-44
US 2002081670 A1 20020627 (200245) C12P019-04
JP 2002519054 W 20020702 (200246) 127 C12N015-09
MX 2000012491 A1 20030701 (200366) C12N000-00000
US 2003190738 A1 20031009 (200367) C12P019-04
IN BISGARD-FRANTZEN, H; SVENDSEN, A

L13 ANSWER 58 OF 184 MEDLINE on STN DUPLICATE 27
TI D-Alanine substitution of teichoic acids as a modulator of protein folding and **stability** at the cytoplasmic membrane/cell wall interface of **Bacillus** subtilis.
SO Journal of biological chemistry, (2000 Sep 1) 275 (35) 26696-703.
Journal code: 2985121R. ISSN: 0021-9258.
AU Hyyrylainen H L; Vitikainen M; Thwaite J; Wu H; Sarvas M; Harwood C R; Kontinen V P; Stephenson K
AN 2000472614 MEDLINE

L13 ANSWER 59 OF 184 MEDLINE on STN DUPLICATE 28
TI Probing structural determinants specifying high **thermostability** in **Bacillus** licheniformis **alpha-amylase.**
SO Journal of molecular biology, (2000 Aug 25) 301 (4) 1041-57.
Journal code: 2985088R. ISSN: 0022-2836.
AU Declerck N; Machius M; Wiegand G; Huber R; Gaillardin C
AN 2000438427 MEDLINE

L13 ANSWER 60 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI Protein engineering of new industrial amylases
SO TRENDS IN GLYCOSCIENCE AND GLYCOTECHNOLOGY, (NOV 2000) Vol. 12, No. 68, pp. 389-401.
Publisher: FCCA-FORUM CARBOHYDRATES COMING AGE, C/O GAKUSHIN PUBLISHING CO LTD 1-1-8 TARUMI-CHO, SUITA 564-0062, OSAKA, 30015, JAPAN.
ISSN: 0915-7352.
AU Hashida M (Reprint); Bisgaard-Frantzen H
AN 2001:215308 SCISEARCH

L13 ANSWER 61 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI Development of marker-free strains of **Bacillus** subtilis capable of secreting high levels of industrial enzymes
SO JOURNAL OF INDUSTRIAL MICROBIOLOGY & BIOTECHNOLOGY, (OCT 2000) Vol. 25, No. 4, pp. 204-212.
Publisher: NATURE AMERICA INC, 345 PARK AVE SOUTH, NEW YORK, NY 10010-1707 USA.
ISSN: 1367-5435.
AU Widner B (Reprint); Thomas M; Sternberg D; Lammon D; Behr R; Sloma A

AN 2001:121629 SCISEARCH

L13 ANSWER 62 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Structure of β-amylase: X-ray crystallographic analysis
SO Glycoenzymes (2000), 55-81. Editor(s): Ohnishi, Masatake. Publisher: Japan Scientific Societies Press, Tokyo, Japan.
CODEN: 69AQDK
AU Mikami, Bunzo
AN 2000:806067 HCAPLUS
DN 133:331218

L13 ANSWER 63 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI New termamyl-like **alpha-amylase variants**; recombinant enzyme production via vector plasmid pTVB106-mediated gene transfer and expression in **Bacillus** subtilis for enzyme stabilization and use in the food industry
AU Borchert T V; Svendsen A; Andersen C; Nielsen B R; Nissen T L; Kjaerulff S
AN 1999-10209 BIOTECHDS
PI WO 9923211 14 May 1999 .

L13 ANSWER 64 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 30
TI Maltogenic α **-amylase variants** with altered properties
SO PCT Int. Appl., 146 pp.
CODEN: PIXXD2
IN Cherry, Joel Robert; Svendsen, Allan; Andersen, Carsten; Beier, Lars; Frandsen, Torben Peter
AN 1999:566161 HCAPLUS
DN 131:181666

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9943794	A1	19990902	WO 1999-DK88	19990226
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
CA 2321595	AA	19990902	CA 1999-2321595	19990226
AU 9925129	A1	19990915	AU 1999-25129	19990226
AU 757935	B2	20030313		
BR 9908281	A	20001031	BR 1999-8281	19990226
EP 1058724	A1	20001213	EP 1999-904736	19990226
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, NL, SE, PT, IE, FI				
NZ 505820	A	20021025	NZ 1999-505820	19990226
JP 2003521866	T2	20030722	JP 2000-533534	19990226
US 6162628	A	20001219	US 1999-386607	19990831

L13 ANSWER 65 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 31
TI **Mutant Bacillus** licheniformis **alpha-amylase** with improved low pH performance and their use in starch liquefaction and in detergents
SO PCT Int. Appl., 38 pp.
CODEN: PIXXD2
IN Caldwell, Robert M.; Mitchinson, Colin; Ropp, Traci H.
AN 1999:388308 HCAPLUS
DN 131:41524

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9929876	A2	19990617	WO 1998-US25124	19981201

WO 9929876 A3 19990722
W: AL, AM, AT, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, CZ, DE, DE, DK, DK, EE, EE, ES, FI, FI, GB, GE, GH, GM, HR, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
US 6211134 B1 20010403 US 1997-985659 19971209
CA 2312053 AA 19990617 CA 1998-2312053 19981201
AU 9916038 A1 19990628 AU 1999-16038 19981201
EP 1038007 A2 20000927 EP 1998-960453 19981201
R: AT, BE, CH, DE, DK, ES, FR, GB, IT, LI, NL, FI
JP 2001526038 T2 20011218 JP 2000-524447 19981201

L13 ANSWER 66 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 32
TI α -**Amylase mutants** with improved **thermostability** for use as detergent additives and for starch liquefaction
SO PCT Int. Appl., 93 pp.
CODEN: PIXXD2
IN Svendsen, Allan; Borchert, Torben Vedel; Bisgard-Frantzen, Henrik
AN 1999:271480 HCAPLUS
DN 130:308445

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9919467	A1	19990422	WO 1998-DK444	19981013
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
CA 2305191	AA	19990422	CA 1998-2305191	19981013
AU 9894343	A1	19990503	AU 1998-94343	19981013
EP 1023439	A1	20000802	EP 1998-947417	19981013
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, NL, SE, PT, IE, FI				
JP 2001520006	T2	20011030	JP 2000-516020	19981013

L13 ANSWER 67 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI New **mutant Bacillus** licheniformis **alpha-amylase**; **mutant** enzyme production and characterization for used in the food and textile industry
AU Day A; Swanson B
AN 1999-06459 BIOTECHDS
PI WO 9909183 25 Feb 1999

L13 ANSWER 68 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Recombinant disulfide-linked **mutant** α - **amylase** with improved **stability** for use in detergents and starch liquefaction
SO PCT Int. Appl., 35 pp.
CODEN: PIXXD2
IN Day, Anthony G.
AN 1999:64948 HCAPLUS
DN 130:135889

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9902702	A1	19990121	WO 1998-US13572	19980629

W: AL, AM, AT, AU, AZ, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, UZ, VN, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG

US 6008026 A 19991228 US 1997-890383 19970711
AU 9884738 A1 19990208 AU 1998-84738 19980629
EP 1002098 A1 20000524 EP 1998-935504 19980629
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI
JP 2001509389 T2 20010724 JP 2000-502196 19980629
MX 200000384 A 20001020 MX 2000-384 20000110

L13 ANSWER 69 OF 184 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI New **variants** of maltogenic **alpha-amylase** or cyclodextrin glucanotransferase and their hybrids, used as anti-staling additives for bread and for production of cyclodextrins.
PI WO 9943793 A1 19990902 (199945)* EN 58 C12N009-28
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL OA PT SD SE SL SZ UG ZW
W: AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT UA UG US UZ VN YU ZW
AU 9925128 A 19990915 (200004) C12N009-28
EP 1066374 A1 20010110 (200103) EN C12N009-28
R: AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE
CN 1292028 A 20010418 (200141) C12N009-28
US 6482622 B1 20021119 (200280) C12N009-00
US 2003059902 A1 20030327 (200325) C12P019-04
AU 761751 B 20030612 (200349) C12N009-28
US 2003207408 A1 20031106 (200374) A21D008-02
US 2003215928 A1 20031120 (200377) C12P019-18
IN ANDERSEN, C; BEIER, L; CHERRY, J R; FRANDSEN, T P; SCHAEFER, T; SVENDSEN, A; ANDERSEN, C D; SCHAFER, T

L13 ANSWER 70 OF 184 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI A Microdochium nivale carbohydrate oxidase and related polynucleotide sequence.
PI WO 9931990 A1 19990701 (199933)* EN 80 A21D008-04
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL OA PT SD SE SZ UG ZW
W: AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT UA UG UZ VN YU ZW
AU 9917518 A 19990712 (199950) A21D008-04
EP 1041890 A1 20001011 (200052) EN A21D008-04
R: AT BE CH DE DK ES FI FR GB GR IE IT LI NL PT SE
US 6165761 A 20001226 (200103) C12N009-04
CN 1283082 A 20010207 (200129) A21D008-04
JP 2001526058 W 20011218 (200203) 96 C12N015-09
AU 753578 B 20021024 (200277) A21D008-04
CN 1379989 A 20021120 (200319) A21D008-04
US 2003180416 A1 20030925 (200364) A21D008-02
IN CHRISTENSEN, S; DYBDAL, L; FUGLSANG, C C; GOLIGHTLY, E; SCHNEIDER, P; XU, F

L13 ANSWER 71 OF 184 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI Cleaning compositions used in e.g. detergent for cleaning hard surfaces or fabrics, dishwashing or oral cleaning comprises protease and amylase

variants having amino acid residues.

PI WO 9920723 A2 19990429 (199934)* EN 168 C11D000-00
RW: AT BE CH CY DE DK EA ES FI FR GB GH GM GR IE IT KE LS LU MC MW NL OA PT SD SE SZ UG ZW
W: AL AM AT AU AZ BA BB BG BR BY CA CH CN CU CZ DE DK EE ES FI GB GD GE GH GM HR HU ID IL IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT UA UG US UZ VN YU ZW
AU 9911971 A 19990510 (199938)
EP 1082404 A2 20010314 (200116) EN C11D001-00
R: AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU NL PT SE
BR 9815230 A 20011002 (200167) C12N009-54
JP 2001520305 W 20011030 (200202) 211 C11D003-386
CZ 2000001478 A3 20011212 (200206) C11D003-386
AU 742632 B 20020110 (200217) C11D003-386
HU 2001004539 A2 20020429 (200238) C11D003-386

IN BAECK, A C; BUSCH, A; GHOSH, C K; OHTANI, R; SHOWELL, M S

L13 ANSWER 72 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI A unique chitinase with dual active sites and triple substrate binding sites from the hyperthermophilic archaeon Pyrococcus kodakaraensis KOD1
SO APPLIED AND ENVIRONMENTAL MICROBIOLOGY, (DEC 1999) Vol. 65, No. 12, pp. 5338-5344.
Publisher: AMER SOC MICROBIOLOGY, 1325 MASSACHUSETTS AVENUE, NW, WASHINGTON, DC 20005-4171.
ISSN: 0099-2240.
AU Tanaka T; Fujiwara S; Nishikori S; Fukui T; Takagi M; Imanaka T (Reprint)
AN 1999:949061 SCISEARCH

L13 ANSWER 73 OF 184 MEDLINE on STN
TI Crystal structure of Thermoactinomyces vulgaris R-47 **alpha-amylase** II (TVAII) hydrolyzing cyclodextrins and pullulan at 2.6 A resolution.
SO Journal of molecular biology, (1999 Apr 16) 287 (5) 907-21.
Journal code: 2985088R. ISSN: 0022-2836.
AU Kamitori S; Kondo S; Okuyama K; Yokota T; Shimura Y; Tonozuka T; Sakano Y
AN 1999241045 MEDLINE

L13 ANSWER 74 OF 184 MEDLINE on STN
TI Electrostatics in the active site of an **alpha-amylase.**
SO European journal of biochemistry / FEBS, (1999 Sep) 264 (3) 816-24.
Journal code: 0107600. ISSN: 0014-2956.
AU Nielsen J E; Beier L; Otzen D; Borchert T V; Frantzen H B; Andersen K V; Svendsen A
AN 1999421687 MEDLINE

L13 ANSWER 75 OF 184 MEDLINE on STN DUPLICATE 33
TI Protein engineering of **alpha-amylase** for low pH performance.
SO Current opinion in biotechnology, (1999 Aug) 10 (4) 349-52. Ref: 14
Journal code: 9100492. ISSN: 0958-1669.
AU Shaw A; Bott R; Day A G
AN 1999380788 MEDLINE

L13 ANSWER 76 OF 184 Elsevier BIOBASE COPYRIGHT 2004 Elsevier Science B.V. on STN
AN 1999117563 ESBIOBASE
TI Expression of **Bacillus** macerans cyclodextrin glucanotransferase in **Bacillus** subtilis
AU Kim C.-S.; Nam Soo Han; Kweon D.-H.; Seo J.-H.
CS J.-H. Seo, Dept. of Food Science and Technology, Res. Ctr. for New Bio-Mat. in Agric., Seoul National University, Suwon 441-744, South Korea.
E-mail: jhseo94@plaza.snu.ac.kr

SO Journal of Microbiology and Biotechnology, (1999), 9/2 (230-233), 15 reference(s)
CODEN: JOMBES ISSN: 1017-7825
DT Journal; Article
CY Korea, Republic of
LA English
SL English

L13 ANSWER 77 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Use of specific **alpha-amylase** enzymes;
enzyme engineering for application in laundry surfactant composition
AU Baeck A C; Jones L A; Ohtani R; Pramod K; Rai S; Showell M S
AN 1998-05749 BIOTECHDS
PI WO 9805748 12 Feb 1998

L13 ANSWER 78 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Starch liquefaction;
using **Bacillus** licheniformis **alpha-amylase**
mutant with improved oxidative **stability**
AU Barnett C C; Solheim L P; Mitchinson C; Power S D; Requadt C A
AN 1999-03126 BIOTECHDS
PI US 5849549 15 Dec 1998

L13 ANSWER 79 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Metallo-endopeptidases with improved **stability**, their manufacture with recombinant cells, and their industrial use
SO PCT Int. Appl., 41 pp.
CODEN: PIXXD2
IN Van Den Burg, Lambertus; Veltman, Oene Robert; Venema, Gerard
AN 1998:684976 HCAPLUS
DN 129:286734

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9844127	A1	19981008	WO 1998-NL164	19980323
W: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, GW, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SZ, UG, ZW, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG				
AU 9865259	A1	19981022	AU 1998-65259	19980323
EP 970225	A1	20000112	EP 1998-911274	19980323
R: BE, CH, DE, DK, FR, GB, IT, LI, NL, IE, FI				
US 6518054	B1	20030211	US 1999-381982	19991207

L13 ANSWER 80 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Pullulanase **mutants** of **Bacillus** strain KSM-AP1378 for preparation of detergents and starch-saccharifying agents
SO Jpn. Kokai Tokkyo Koho, 19 pp.
CODEN: JKXXAF
IN Sumitomo, Nobuyuki; Hatada, Yuji; Ichimura, Takashi; Saito, Kazuhiro; Kawai, Shuji; Ito, Susumu
AN 1998:794818 HCAPLUS
DN 130:106926

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI JP 10327868	A2	19981215	JP 1997-141596	19970530

L13 ANSWER 81 OF 184 WPIDS COPYRIGHT 2004 THOMSON DERWENT on STN
TI DNA encoding **mutant** and **variant alpha-amylase** proteins - of **Bacillus** licheniformis, useful for producing recombinant **alpha-amylase** proteins.

PI US 5824532 A 19981020 (199849)* 56 C12N015-56
IN BARNETT, C C; MITCHINSON, C; POWER, S D; REQUADT, C A

L13 ANSWER 82 OF 184 MEDLINE on STN DUPLICATE 36
TI Engineering of cyclodextrin product specificity and pH optima of the thermostable cyclodextrin glycosyltransferase from Thermoanaerobacterium thermosulfurigenes EM1.
SO Journal of biological chemistry, (1998 Mar 6) 273 (10) 5771-9.
Journal code: 2985121R. ISSN: 0021-9258.
AU Wind R D; Uitdehaag J C; Buitelaar R M; Dijkstra B W; Dijkhuizen L
AN 1998157977 MEDLINE

L13 ANSWER 83 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI Enzymatic properties of a novel liquefying **alpha-amylase** from an alkaliphilic **Bacillus** isolate and entire nucleotide and amino acid sequences
SO APPLIED AND ENVIRONMENTAL MICROBIOLOGY, (SEP 1998) Vol. 64, No. 9, pp. 3282-3289.
Publisher: AMER SOC MICROBIOLOGY, 1325 MASSACHUSETTS AVENUE, NW, WASHINGTON, DC 20005-4171.
ISSN: 0099-2240.
AU Igarashi K; Hatada Y; Hagihara H; Saeki K; Takaiwa M; Uemura T; Ara K; Ozaki K; Kawai S; Kobayashi T; Ito S (Reprint)
AN 1998:713242 SCISEARCH

L13 ANSWER 84 OF 184 MEDLINE on STN DUPLICATE 37
TI Crystal structure of a catalytic-site **mutant alpha-amylase** from **Bacillus** subtilis complexed with maltopentaose.
SO Journal of molecular biology, (1998 Mar 27) 277 (2) 393-407.
Journal code: 2985088R. ISSN: 0022-2836.
AU Fujimoto Z; Takase K; Doui N; Momma M; Matsumoto T; Mizuno H
AN 1998181035 MEDLINE

L13 ANSWER 85 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 38
TI An Escherichia coli host strain useful for efficient overproduction of secreted recombinant protein
SO BIOTECHNOLOGY AND BIOENGINEERING, (5 AUG 1998) Vol. 59, No. 3, pp. 386-391.
Publisher: JOHN WILEY & SONS INC, 605 THIRD AVE, NEW YORK, NY 10158-0012.
ISSN: 0006-3592.
AU Weikert C; Sauer U; Bailey J E (Reprint)
AN 1998:498059 SCISEARCH

L13 ANSWER 86 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Improved **thermostability** of a **Bacillus .alpha .-amylase** by deletion of an arginine-glycine residue is caused by enhanced **calcium** binding
SO Biochemical and Biophysical Research Communications (1998), 248(2), 372-377
CODEN: BBRCA9; ISSN: 0006-291X
AU Igarashi, Kazuaki; Hatada, Yuji; Ikawa, Kaori; Araki, Hiroyuki; Ozawa, Tadahiro; Kobayashi, Tohru; Ozaki, Katsuya; Ito, Susumu
AN 1998:493007 HCAPLUS
DN 129:213459

L13 ANSWER 87 OF 184 MEDLINE on STN DUPLICATE 39
TI Activation of **Bacillus** licheniformis **alpha-amylase** through a disorder-->order transition of the substrate-binding site mediated by a **calcium**-sodium-**calcium** metal triad.
SO Structure (London, England), (1998 Mar 15) 6 (3) 281-92.
Journal code: 9418985. ISSN: 0969-2126.

AU Machius M; Declerck N; Huber R; Wiegand G
AN 1998212915 MEDLINE

L13 ANSWER 88 OF 184 Elsevier BIOBASE COPYRIGHT 2004 Elsevier Science B.V. on STN DUPLICATE
AN 1998079486 ESBIOBASE
TI Activation of **Bacillus** licheniformis α - **amylase** through a disorder → order transition of the substrate-binding site mediated by a **calcium**-sodium-**calcium** metal triad
AU Machius M.; Declerck N.; Huber R.; Wiegand G.
CS M. Machius, Max-Planck-Institut fur Biochemie, D-85152 Planegg-Martinsried, Germany.
E-mail: machius@chop.swmed.edu
SO Structure, (15 MAR 1998), 6/3 (281), 45 reference(s)
CODEN: STRUE6 ISSN: 0969-2126
DT Journal; Article
CY United Kingdom
LA English
SL English

L13 ANSWER 89 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI Protein thermostabilization by proline substitutions
SO JOURNAL OF MOLECULAR CATALYSIS B-ENZYMATIC, (14 JUN 1998) Vol. 4, No. 4, pp. 167-180.
Publisher: ELSEVIER SCIENCE BV, PO BOX 211, 1000 AE AMSTERDAM, NETHERLANDS.
ISSN: 1381-1177.
AU Watanabe K; Suzuki Y (Reprint)
AN 1998:536593 SCISEARCH

L13 ANSWER 90 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Hyperthermostable extracellular **alpha-amylase** from Pyrococcus furiosus;
thermophilic bacterium recombinant enzyme production and characterization (conference abstract)
SO Abstr.Pap.Am.Chem.Soc.; (1998) 216 Meet. Pt.3, BTEC019
CODEN: ACSRAL ISSN: 0065-7727
216th ACS National Meeting, Boston, MA, USA, 23-27 August, 1998, 216 Meet., Pt.3, 1998.
AU Savchenko A; Dong G; Vieille C; Zeikus G J
AN 1999-14174 BIOTECHDS

L13 ANSWER 91 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Termamyl-like **alpha-amylase variants** with improved properties;
enzyme engineering and expression in **Bacillus** spp.
AU Svensden A; Borchert T V; Bisgard-Frantzen H
AN 1998-01800 BIOTECHDS
PI WO 9741213 6 Nov 1997

L13 ANSWER 92 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Detergent compositions for hard surface cleaning and laundry use;
Bacillus sp. **alpha-amylase**-containing surfactant composition
AU Baeck A C; Jones L A; Ohtani R; Pramod K; Raj S; Showell M S; Ward G
AN 1997-12476 BIOTECHDS
PI WO 9732961 12 Sep 1997

L13 ANSWER 93 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI New modified **alpha-amylase** enzymes;
enzyme engineering
AU Bott R R; Shaw A
AN 1998-02380 BIOTECHDS

PI WO 9743424 20 Nov 1997

L13 ANSWER 94 OF 184 MEDLINE on STN DUPLICATE 43
TI Hyperthermostable **mutants** of **Bacillus** licheniformis **alpha-amylase**: thermodynamic studies and structural interpretation.
SO Protein engineering, (1997 May) 10 (5) 541-9.
Journal code: 8801484. ISSN: 0269-2139.
AU Declerck N; Machius M; Chambert R; Wiegand G; Huber R; Gaillardin C
AN 97358476 MEDLINE

L13 ANSWER 95 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Strain improvement for the production of a thermostable **alpha-amylase**;
Bacillus stearothermophilus mutagenesis, and gene cloning and expression in Escherichia coli and **Bacillus** subtilis
SO Enzyme Microb.Technol.; (1997) 21, 7, 525-30
CODEN: EMTED2 ISSN: 0141-0229
AU Sidhu G S; Sharma P; Chakrabarti T; *Gupta J K
AN 1998-00332 BIOTECHDS

L13 ANSWER 96 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 44
TI Purification, characterisation and mutagenic enhancement of a thermoactive **alpha-amylase** from **Bacillus** subtilis
SO JOURNAL OF INDUSTRIAL MICROBIOLOGY & BIOTECHNOLOGY, (OCT 1997) Vol. 19, No. 4, pp. 273-279.
Publisher: STOCKTON PRESS, HOUNDMILLS, BASINGSTOKE, HAMPSHIRE, ENGLAND RG21 6XS.
ISSN: 0169-4146.
AU Uguru G C (Reprint); Robb D A; Akinyanju J A; Sani A
AN 1998:7966 SCISEARCH

L13 ANSWER 97 OF 184 Elsevier BIOBASE COPYRIGHT 2004 Elsevier Science B.V. on STN
AN 1997167032 ESBIOBASE
TI Instability of α -**amylase** production and morphological variation in continuous culture of **Bacillus** amyloliquefaciens is associated with plasmid loss
AU Hillier P.; Wase D.A.J.; Emery A.N.; Solomons G.L.
CS P. Hillier, School of Chemical Engineering, University of Birmingham, P.O. Box 363, Edgbaston, Birmingham B15 2TT, United Kingdom.
SO Process Biochemistry, (1997), 32/1 (51-59), 19 reference(s)
CODEN: PBCHE5 ISSN: 0032-9592
PUI S0032959296000489
DT Journal; Article
CY United Kingdom
LA English
SL English

L13 ANSWER 98 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI New **alpha-amylase variants**;
mutant enzyme construction for improved **calcium** dependency, substrate binding, cleavage, pH dependent activity and **thermostability**; application in e.g. surfactant composition
AU Svendsen A; Bisgard-Frantzen H; Borchert T V
AN 1996-12567 BIOTECHDS
PI WO 9623874 8 Aug 1996

L13 ANSWER 99 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI New **alpha-amylase variants**;
recombinant vector expression in bacterium or fungus for **mutant** enzyme production; application in surfactant composition etc.

AU Bisgard-Frantzen H; Svendsen A; Borchert T V
AN 1996-12566 BIOTECHDS
PI WO 9623873 8 Aug 1996

L13 ANSWER 100 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN DUPLICATE 47
TI An improved laundry detergent composition containing amylase **mutants**
SO PCT Int. Appl., 105 pp.
CODEN: PIXXD2
IN Barnett, Christopher C.; Boyer, Stephen G.; Mitchinson, Colin; Power, Scott D.
AN 1996:694369 HCAPLUS
DN 125:303862

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9630481	A1	19961003	WO 1996-US4029	19960322
W: AU, BR, CA, CN, CZ, FI, HU, JP, KR, MX, NO, NZ, PL, RO, RU, VN				
RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
AU 9653226	A1	19961016	AU 1996-53226	19960322
AU 718509	B2	20000413		
EP 815193	A1	19980107	EP 1996-909854	19960322
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, NL, SE, PT, IE, FI				
CN 1179176	A	19980415	CN 1996-192801	19960322
BR 9607751	A	19980623	BR 1996-7751	19960322
JP 11502562	T2	19990302	JP 1996-529561	19960322
NO 9704402	A	19971119	NO 1997-4402	19970923

L13 ANSWER 101 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI **Bacillus** α **-amylase mutant** recombinant production, improved low pH starch liquefaction, thermal **stability**, and activity, and use as laundry detergent or dishwashing detergent
SO PCT Int. Appl., 48 pp.
CODEN: PIXXD2
IN Mitchinson, Colin; Requadt, Carol; Ropp, Traci; Solheim, Leif P.; Ringer, Christopher; Day, Anthony
AN 1997:88800 HCAPLUS
DN 126:105762

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9639528	A2	19961212	WO 1996-US9089	19960606
WO 9639528	A3	19970213		
W: AL, AM, AT, AU, AZ, BB, BG, BR, BY, CA, CH, CN, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IS, JP, KE, KG, KP, KR, KZ, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI				
RW: KE, LS, MW, SD, SZ, UG, AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN				
US 5736499	A	19980407	US 1995-468700	19950606
CA 2222726	AA	19961212	CA 1996-2222726	19960606
AU 9662557	A1	19961224	AU 1996-62557	19960606
EP 832250	A2	19980401	EP 1996-921305	19960606
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, NL, SE, PT, IE, FI				
CN 1191570	A	19980826	CN 1996-195005	19960606
CN 1111601	B	20030618		
BR 9608647	A	19990504	BR 1996-8647	19960606
JP 11506941	T2	19990622	JP 1996-501492	19960606
US 5958739	A	19990928	US 1997-704706	19970220

L13 ANSWER 102 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI An improved cleaning composition containing **Bacillus** licheniformis α **-amylase mutants** with improved thermal **stability** and oxidation resistance
SO PCT Int. Appl., 85 pp.

CODEN: PIXXD2
IN Barnett, Christopher C.; Mitchinson, Colin; Power, Scott D.
AN 1996:323628 HCAPLUS
DN 125:4407

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI WO 9605295	A2	19960222	WO 1995-US10426	19950809
WO 9605295	A3	19960328		
W: AU, BR, CA, CN, CZ, FI, HU, JP, KR, MX, NO, NZ, PL, RU, VN				
RW: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
CA 2197203	AA	19960222	CA 1995-2197203	19950809
AU 9533662	A1	19960307	AU 1995-33662	19950809
AU 686007	B2	19980129		
EP 775201	A2	19970528	EP 1995-930186	19950809
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LI, LU, MC, NL, PT, SE				
CN 1158637	A	19970903	CN 1995-194852	19950809
JP 10504197	T2	19980428	JP 1995-507603	19950809
BR 9508582	A	19980602	BR 1995-8582	19950809
HU 77748	A2	19980728	HU 1998-643	19950809
FI 9700563	A	19970210	FI 1997-563	19970210
NO 9700609	A	19970324	NO 1997-609	19970210

L13 ANSWER 103 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI RAW-STARCH-DIGESTING AND THERMOSTABLE **ALPHA-AMYLASE** FROM THE YEAST CRYPTOCOCCUS SP. S-2 - PURIFICATION, CHARACTERIZATION, CLONING AND SEQUENCING
SO BIOCHEMICAL JOURNAL, (15 SEP 1996) Vol. 318, Part 3, pp. 989-996. ISSN: 0264-6021.
AU IEFUJI H (Reprint); CHINO M; KATO M; IIMURA Y
AN 96:716857 SCISEARCH

L13 ANSWER 104 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI ANALYSIS OF PROTEIN CONFORMATIONAL CHARACTERISTICS RELATED TO **THERMOSTABILITY**
SO PROTEIN ENGINEERING, (MAR 1996) Vol. 9, No. 3, pp. 265-271. ISSN: 0269-2139.
AU QUEROL E; PEREZPONS J A; MOZOVILLARIAS A (Reprint)
AN 96:417859 SCISEARCH

L13 ANSWER 105 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Hyperthermostable **mutants** of **Bacillus** licheniformis: thermodynamic studies and structural interpretation
SO Perspectives on Protein Engineering '96, [International Conference], 5th, Montpellier, Fr., 1996 (1996), Paper No. 7, 9 pp.. Editor(s): Geisow, Michael J. Publisher: BIODIGM, Bingham, UK. CODEN: 64HIAR
AU Declerck, Nathalie; Gaillardin, Claude; Machius, Mischa; Wiegand, Georg; Huber, Robert
AN 1997:287296 HCAPLUS
DN 126:314064

L13 ANSWER 106 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI **Mutant** B. licheniformis **alpha-amylase** enzymes;
Bacillus licheniformis **mutant** thermostable enzyme production; application in starch degradation, textile or paper desizing, brewing industry and as household surfactant
AU van der Laan J M; Aehle W
AN 1996-03039 BIOTECHDS
PI WO 9535382 28 Dec 1995

L13 ANSWER 107 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI New **alpha-amylase variants**;
Bacillus liquefaciens **alpha-amylase**

enzyme engineering for improved **thermostability**, pH **stability**, etc.; application in surfactant composition to improve washing performance
AU Bisgard-Frantzen H; Borchert T V; Svendsen A; Thellersen M; van der Zee P
AN 1995-07973 BIOTECHDS
PI WO 9510603 20 Apr 1995

L13 ANSWER 108 OF 184 MEDLINE on STN DUPLICATE 50
TI Hyperthermostable **mutants** of **Bacillus** licheniformis **alpha-amylase**: multiple amino acid replacements and molecular modelling.
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Journal code: 8801484. ISSN: 0269-2139.
AU Declerck N; Joyet P; Trosset J Y; Garnier J; Gaillardin C
AN 96367070 MEDLINE

L13 ANSWER 109 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI **BACILLUS**-SUBTILIS LEVANSUCRASE - THE EFFICIENCY OF THE 2ND STAGE OF SECRETION IS MODULATED BY EXTERNAL EFFECTORS ASSISTING FOLDING
SO MICROBIOLOGY-UK, (APR 1995) Vol. 141, Part 4, pp. 997-1005.
ISSN: 1350-0872.
AU CHAMBERT R (Reprint); HADDAOUI E A; PETITGLATRON M F
AN 95:296718 SCISEARCH

L13 ANSWER 110 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 51
TI **THERMOSTABILITY** OF **ALPHA-AMYLASE** PRODUCED BY **BACILLUS** SP E2 - A THERMOPHILIC **MUTANT**
SO WORLD JOURNAL OF MICROBIOLOGY & BIOTECHNOLOGY, (SEP 1995) Vol. 11, No. 5, pp. 593-594.
ISSN: 0959-3993.
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AN 95:679350 SCISEARCH

L13 ANSWER 111 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI **Thermostability** of **alpha-amylase** produced by **Bacillus** sp. E2 - a thermophilic **mutant**; enzyme characterization produced by thermophilic bacterium
SO World J.Microbiol.Biotechnol.; (1995) 11, 5, 593-94
CODEN: 9295H ISSN: 0959-3993
AU Goyal N; Sidhu G S; Chakrabarti T; *Gupta J K
AN 1995-14132 BIOTECHDS

L13 ANSWER 112 OF 184 MEDLINE on STN DUPLICATE 52
TI Co-overexpression of prlF increases cell viability and enzyme yields in recombinant Escherichia coli expressing **Bacillus** stearothermophilus **alpha-amylase.**
SO Biotechnology progress, (1995 Jul-Aug) 11 (4) 403-11.
Journal code: 8506292. ISSN: 8756-7938.
AU Minas W; Bailey J E
AN 95382886 MEDLINE

L13 ANSWER 113 OF 184 Elsevier BIOBASE COPYRIGHT 2004 Elsevier Science B.V. on STN
AN 1995126737 ESBIOBASE
TI Colony switching in an **alpha-amylase**-producing strain of **Bacillus** subtilis
AU Rodriguez H.
CS H. Rodriguez, Department of Microbiology, Cuban Res. Inst. Sugarcane By-prod., (ICIDCA), PO Box 4026, CP 11 000 C Habana, Cuba.
SO Journal of Industrial Microbiology, (1995), 15/2 (112-115)
CODEN: JIMIE7 ISSN: 0169-4146
DT Journal; Article
CY United Kingdom

LA English
SL English

L13 ANSWER 114 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI THE ROLE OF HISTIDINE-RESIDUES IN THE CATALYTIC ACT OF CYCLOMALTODEXTRIN GLUCANOTRANSFERASE FROM **BACILLUS**-CIRCULANS VAR ALKALOPHILUS
SO BIOCHIMICA ET BIOPHYSICA ACTA-PROTEIN STRUCTURE AND MOLECULAR ENZYMOLOGY, (22 FEB 1995) Vol. 1247, No. 1, pp. 97-103.
ISSN: 0167-4838.
AU MATTSSON P (Reprint); BATTCHIKOVA N; SIPPOLA K; KORPELA T
AN 95:163838 SCISEARCH

L13 ANSWER 115 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI SURVIVAL OF **BACILLUS**-SUBTILIS NB22 AND ITS TRANSFORMANT IN SOIL
SO APPLIED SOIL ECOLOGY, (JUN 1995) Vol. 2, No. 2, pp. 85-94.
ISSN: 0929-1393.
AU TOKUDA Y; ANO T; SHODA M (Reprint)
AN 95:519809 SCISEARCH

L13 ANSWER 116 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI **Bacillus** licheniformis, **Bacillus** stearothermophilus and **Bacillus** amyloliquefaciens **alpha-amylase** enzyme engineering by site-directed mutagenesis; DNA sequence; application in a surfactant or a starch liquefaction composition
AN 1994-13784 BIOTECHDS
PI WO 9418314 18 Aug 1994

L13 ANSWER 117 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI **Mutant alpha-amylase** from **Bacillus** sp. use as surfactant, dish washing agent and liquefaction agent; **Bacillus** or Aspergillus spp. thermostable enzyme with increased **thermostability** and activity at low pH produced by enzyme engineering
AN 1994-04189 BIOTECHDS
PI WO 9402597 3 Feb 1994

L13 ANSWER 118 OF 184 MEDLINE on STN DUPLICATE 55
TI Four aromatic residues in the active center of cyclodextrin glucanotransferase from alkalophilic **Bacillus** sp. 1011: effects of replacements on substrate binding and cyclization characteristics.
SO Biochemistry, (1994 Aug 23) 33 (33) 9929-36.
Journal code: 0370623. ISSN: 0006-2960.
AU Nakamura A; Haga K; Yamane K
AN 94339126 MEDLINE

L13 ANSWER 119 OF 184 MEDLINE on STN DUPLICATE 56
TI C-terminal truncations of a thermostable **Bacillus** stearothermophilus **alpha-amylase.**
SO Protein engineering, (1994 Oct) 7 (10) 1255-9.
Journal code: 8801484. ISSN: 0269-2139.
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AN 95158398 MEDLINE

L13 ANSWER 120 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI RANDOM MUTAGENESIS OF PULLULANASE FROM KLEBSIELLA-AEROGENES FOR STUDIES OF THE STRUCTURE AND FUNCTION OF THE ENZYME
SO JOURNAL OF BIOCHEMISTRY, (DEC 1994) Vol. 116, No. 6, pp. 1233-1240.
ISSN: 0021-924X.
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AN 95:3862 SCISEARCH

L13 ANSWER 121 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 57

TI CHANGES IN OPTIMUM PH AND **THERMOSTABILITY** OF **ALPHA-AMYLASE** FROM **BACILLUS**-LICHENIFORMIS BY SITE-DIRECTED MUTAGENESIS OF HIS-235 AND ASP-328
SO BULLETIN OF THE KOREAN CHEMICAL SOCIETY, (20 OCT 1994) Vol. 15, No. 10, pp. 832-835.
ISSN: 0253-2964.
AU KIM M S (Reprint); LEE S K; JUNG H S; YANG C H
AN 94:725048 SCISEARCH

L13 ANSWER 122 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI RESIDUES ESSENTIAL FOR CATALYTIC ACTIVITY OF SOYBEAN BETA-AMYLASE
SO EUROPEAN JOURNAL OF BIOCHEMISTRY, (15 APR 1994) Vol. 221, No. 2, pp. 649-654.
ISSN: 0014-2956.
AU TOTSUKA A; NONG V H; KADOKAWA H; KIM C S; ITOH Y; FUKAZAWA C (Reprint)
AN 94:253736 SCISEARCH

L13 ANSWER 123 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI SITE-DIRECTED MUTAGENESIS OF HISTIDINE-93, ASPARTIC ACID-180, GLUTAMIC ACID-205, HISTIDINE-290, AND ASPARTIC ACID-291 AT THE ACTIVE-SITE AND TRYPTOPHAN-279 AT THE RAW STARCH BINDING-SITE IN BARLEY **ALPHA-AMYLASE** 1
SO JOURNAL OF BIOLOGICAL CHEMISTRY, (25 OCT 1993) Vol. 268, No. 30, pp. 22480-22484.
ISSN: 0021-9258.
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AN 93:656161 SCISEARCH

L13 ANSWER 124 OF 184 MEDLINE on STN
TI Structural requirements of **Bacillus** subtilis **alpha-amylase** signal peptide for efficient processing: in vivo pulse-chase experiments with **mutant** signal peptides.
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Journal code: 2985120R. ISSN: 0021-9193.
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AN 93308100 MEDLINE

L13 ANSWER 125 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI CRYSTALLIZATION AND PRELIMINARY-X-RAY STUDIES OF WILD-TYPE AND CATALYTIC-SITE **MUTANT ALPHA-AMYLASE** FROM **BACILLUS**-SUBTILIS
SO JOURNAL OF MOLECULAR BIOLOGY, (20 DEC 1993) Vol. 234, No. 4, pp. 1282-1283.
ISSN: 0022-2836.
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AN 93:752000 SCISEARCH

L13 ANSWER 126 OF 184 LIFESCI COPYRIGHT 2004 CSA on STN DUPLICATE 58
TI Studies on extracellular thermostable **alpha -amylase** from **Bacillus** licheniformis
SO ACTA MICROBIOL. SIN., (1993) vol. 33, no. 4, pp. 274-279.
ISSN: 0001-6209.
AU Xianliang, Kong; Junying, Wang; Hongtao, Jiang; Liping, Jiang
AN 95:30857 LIFESCI

L13 ANSWER 127 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI **Stability** of industrial enzymes;
enzyme stabilization by chemical modification or enzyme engineering (conference paper)
SO Stud.Org.Chem.; (1993) 47, 111-31
CODEN: 9999T
AU Misset O
AN 1994-05917 BIOTECHDS

L13 ANSWER 128 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI New thermostable forms of **Bacillus** licheniformis **alpha -amylase**;
enzyme engineering by specific amino acid substitutions at positions 133 and or 209, for simultaneous gelation and liquefaction of starch, e.g. in brewing
AN 1993-03609 BIOTECHDS
PI FR 2676456 20 Nov 1992

L13 ANSWER 129 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI New thermostable **alpha-amylase** from **Bacillus** licheniformis;
obtained by enzyme engineering and useful in paper-making, brewing etc. for starch liquefaction
AN 1992-07694 BIOTECHDS
PI FR 2665178 31 Jan 1992

L13 ANSWER 130 OF 184 MEDLINE on STN DUPLICATE 61
TI Hyperthermostable **variants** of a highly thermostable **alpha-amylase.**
SO Bio/technology (Nature Publishing Company), (1992 Dec) 10 (12) 1579-83.
Journal code: 8309273. ISSN: 0733-222X.
AU Joyet P; Declerck N; Gaillardin C
AN 93168398 MEDLINE

L13 ANSWER 131 OF 184 MEDLINE on STN DUPLICATE 62
TI Site-directed mutagenesis of active site residues in **Bacillus** subtilis **alpha-amylase.**
SO Biochimica et biophysica acta, (1992 Apr 17) 1120 (3) 281-8.
Journal code: 0217513. ISSN: 0006-3002.
AU Takase K; Matsumoto T; Mizuno H; Yamane K
AN 92247808 MEDLINE

L13 ANSWER 132 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI INTERACTION OF CATALYTIC-SITE **MUTANTS** OF **BACILLUS** -SUBTILIS **ALPHA-AMYLASE** WITH SUBSTRATES AND ACARBOSE
SO BIOCHIMICA ET BIOPHYSICA ACTA, (21 AUG 1992) Vol. 1122, No. 3, pp. 278-282.
ISSN: 0006-3002.
AU TAKASE K (Reprint)
AN 92:535785 SCISEARCH

L13 ANSWER 133 OF 184 MEDLINE on STN
TI Extracellular enzymes: gene regulation and structure function relationship studies.
SO Biotechnology (Reading, Mass.), (1992) 22 189-217. Ref: 94
Journal code: 8300602. ISSN: 0740-7378.
AU Jarnagin A S; Ferrari E
AN 92369847 MEDLINE

L13 ANSWER 134 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 63
TI EFFICIENT PRODUCTION OF THERMOSTABLE CLOSTRIDIUM-THERMOSULFUROGENES BETA-AMYLASE BY **BACILLUS**-BREVIS
SO JOURNAL OF FERMENTATION AND BIOENGINEERING, (1992) Vol. 73, No. 2, pp. 112-115.
ISSN: 0922-338X.
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AN 92:163427 SCISEARCH

L13 ANSWER 135 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI FUNCTIONAL-RELATIONSHIPS BETWEEN CYCLODEXTRIN GLUCANOTRANSFERASE FROM AN ALKALOPHILIC **BACILLUS** AND **ALPHA-AMYLASES** - SITE-DIRECTED MUTAGENESIS OF THE CONSERVED 2 ASP AND ONE GLU RESIDUES

SO FEBS LETTERS, (13 JAN 1992) Vol. 296, No. 1, pp. 37-40.
ISSN: 0014-5793.
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AN 92:64653 SCISEARCH

L13 ANSWER 136 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Recombinant **mutant** microbial **alpha-amylase**; **Bacillus** licheniformis enzyme engineering by site-directed mutagenesis of DNA sequence for improved **thermostability**, acid **stability**; use in starch saccharification, textile desizing
AN 1991-04746 BIOTECHDS
PI WO 9100353 10 Jan 1991

L13 ANSWER 137 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI A **mutant** enzyme with reduced **stability**; **Bacillus** amyloliquefaciens **alpha-amylase** **mutant** expression in e.g. Escherichia coli, **Bacillus** , Aspergillus spp.; bread improver with reduced **thermostability** during baking; DNA sequence
AN 1991-04156 BIOTECHDS
PI EP 409299 23 Jan 1991

L13 ANSWER 138 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 65
TI AMYLASE, BETA-GLUCANASE AND PROTEASE ACTIVITIES FROM A **MUTANT** OF **BACILLUS**-SUBTILIS
SO STARCH-STARKE, (1991) Vol. 43, No. 10, pp. 403-409.
AU YIN X S (Reprint); LI Y X; STARK J R
AN 91:646649 SCISEARCH

L13 ANSWER 139 OF 184 LIFESCI COPYRIGHT 2004 CSA on STN DUPLICATE 66
TI Production of thermophilic **alpha** -**amylase** using immobilized transformed Escherichia coli by addition of glycine
SO J. FERMENT. BIOENG., (1991) vol. 71, no. 6, pp. 397-402.
ISSN: 0922-338X.
AU Ariga, O.; Andoh, Y.; Fujishita, Y.; Watari, T.; Sano, Y.
AN 95:1207 LIFESCI

L13 ANSWER 140 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI PRODUCTION OF THERMOPHILIC **ALPHA-AMYLASE** USING IMMOBILIZED TRANSFORMED ESCHERICHIA-COLI BY ADDITION OF GLYCINE
SO JOURNAL OF FERMENTATION AND BIOENGINEERING, (1991) Vol. 71, No. 6, pp. 397-402.
AU ARIGA O (Reprint); ANDOH Y; FUJISHITA Y; WATARI T; SANO Y
AN 91:390037 SCISEARCH

L13 ANSWER 141 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI A new **Bacillus** licheniformis **alpha-amylase** capable of low pH liquefaction; starch saccharification using thermostable enzyme
SO Starch; (1991) 43, 9, 355-60
CODEN: STARDD
AU Antrim R L; Solheim B A; Solheim L; Auterinen A L; Cunefare J; Karppelin S
AN 1991-13798 BIOTECHDS

L13 ANSWER 142 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI Isolation of thermostable α -**amylase** hyperproducing **Bacillus** sp. Number 32H417 and some properties of the enzyme
SO Sanop Misaengmul Hakhoechi (1991), 19(2), 122-7
CODEN: SMHAEH; ISSN: 0257-2389
AU Kim, Moo Sung; O, Pyong Su

AN 1993:556049 HCAPLUS
DN 119:156049

L13 ANSWER 143 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
TI α -**Amylase** deletion **mutants** of **Bacillus**, and their gene cloning and expression
SO Jpn. Kokai Tokkyo Koho, 11 pp.
CODEN: JKXXAF
IN Imanaka, Tadayuki; Sai, Kiken; Ken, Masuo
AN 1991:201125 HCAPLUS
DN 114:201125

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
JP 02222687	A2	19900905	JP 1989-43561	19890225

L13 ANSWER 144 OF 184 MEDLINE on STN DUPLICATE 67
TI Use of amber suppressors to investigate the **thermostability** of **Bacillus** licheniformis **alpha-amylase.** Amino acid replacements at 6 histidine residues reveal a critical position at His-133.
SO Journal of biological chemistry, (1990 Sep 15) 265 (26) 15481-8.
Journal code: 2985121R. ISSN: 0021-9258.
AU Declerck N; Joyet P; Gaillardin C; Masson J M
AN 90368748 MEDLINE

L13 ANSWER 145 OF 184 BIOSIS COPYRIGHT 2004 BIOLOGICAL ABSTRACTS INC. on STN
TI A METHOD TO RAPIDLY OBTAIN **BACILLUS**-LICHENIFORMIS **ALPHA AMYLASE VARIANTS** USING A SET OF SUPPRESSOR TRANSFER RNA GENES AND TEST OF THEIR ACTIVITY AND **STABILITY.**
SO Protein Engineering, (1990) Vol. 3, No. 4, pp. 353.
Meeting Info.: MEETING ON PROTEIN ENGINEERING, 1989, KOBE, JAPAN, AUGUST 20-25, 1989. PROTEIN ENG.
CODEN: PRENE9. ISSN: 0269-2139.
AU DECLERCK N [Reprint author]; JOYET P; MASSON J M
AN 1990:403351 BIOSIS

L13 ANSWER 146 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
TI Protein engineering of **alpha-amylase** from **Bacillus** stearothermophilus;
recombinant enzyme expression in **Bacillus** subtilis;
thermostable enzyme engineering by site-directed mutagenesis
(conference abstract)
SO Protein Eng.; (1990) 3, 4, 352
CODEN: PRENE9
AU Mantsala P; Vihinen M; Ollikka P; Niskanen J; Meyer P; Suominen I
AN 1990-06570 BIOTECHDS

L13 ANSWER 147 OF 184 MEDLINE on STN DUPLICATE 68
TI Efficient production of human **alpha-amylase** by a **Bacillus** brevis **mutant.**
SO Applied microbiology and biotechnology, (1990 Dec) 34 (3) 297-302.
Journal code: 8406612. ISSN: 0175-7598.
AU Konishi H; Sato T; Yamagata H; Udaka S
AN 91254745 MEDLINE

L13 ANSWER 148 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
TI EFFICIENT PRODUCTION OF HUMAN **ALPHA-AMYLASE** BY A **BACILLUS**-BREVIS **MUTANT**
SO APPLIED MICROBIOLOGY AND BIOTECHNOLOGY, (1990) Vol. 34, No. 3, pp. 297-302.
AU KONISHI H; SATO T; YAMAGATA H (Reprint); UDAKA S
AN 91:34627 SCISEARCH

L13 ANSWER 149 OF 184 MEDLINE on STN DUPLICATE 69

TI Site-directed mutagenesis of a thermostable **alpha-amylase** from **Bacillus** stearothermophilus: putative role of three conserved residues.
SO Journal of biochemistry, (1990 Feb) 107 (2) 267-72. Journal code: 0376600. ISSN: 0021-924X.
AU Vihinen M; Ollikka P; Niskanen J; Meyer P; Suominen I; Karp M; Holm L; Knowles J; Mantsala P
AN 90299864 MEDLINE

L13 ANSWER 150 OF 184 MEDLINE on STN DUPLICATE 70
TI Random mutagenesis used to probe the structure and function of **Bacillus** stearothermophilus **alpha-amylase.**
SO Protein engineering, (1990 Jan) 3 (3) 181-91. Journal code: 8801484. ISSN: 0269-2139.
AU Holm L; Koivula A K; Lehtovaara P M; Hemminki A; Knowles J K
AN 90231977 MEDLINE

L13 ANSWER 151 OF 184 LIFESCI COPYRIGHT 2004 CSA on STN DUPLICATE 71
TI An economical, ungraded, stabilized and efficient preparation of **alpha -amylase.**
SO J. BIOTECHNOL., (1990) vol. 16, no. 1-2, pp. 97-108.
AU Shah, N.K.; Upadhyay, C.M.; Nehete, P.N.; Kothari, R.M.; Hegde, M.V.
AN 90:51089 LIFESCI

L13 ANSWER 152 OF 184 MEDLINE on STN DUPLICATE 72
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L13 ANSWER 5 OF 184 MEDLINE on STN DUPLICATE 2
AB It is generally assumed that in proteins hydrophobic residues are not favorable at solvent-exposed sites, and that amino acid substitutions on the surface have little effect on protein **thermostability**. Contrary to these assumptions, we have identified hyperthermostable **variants** of **Bacillus** licheniformis **alpha-amylase** (BLA) that result from the incorporation of hydrophobic residues at the surface. Under highly destabilizing conditions, a **variant** combining five stabilizing mutations unfolds 32 times more slowly and at a temperature 13 degrees C higher than the wild-type. Crystal structure analysis at 1.7 A resolution suggests that stabilization is achieved through (a) extension of the concept of increased hydrophobic packing, usually applied to cavities, to surface indentations, (b) introduction of favorable aromatic-aromatic interactions on the surface, (c) specific stabilization of intrinsic metal binding sites, and (d) stabilization of a beta-sheet by introducing a residue with high beta-sheet forming propensity. All mutated residues are involved in forming complex, cooperative interaction networks that extend from the interior of the protein to its surface and which may therefore constitute "weak points" where BLA unfolding is initiated. This might explain the unexpectedly large effect induced by some of the substitutions on the kinetic **stability** of BLA. Our study shows that substantial protein stabilization can be achieved by stabilizing surface positions that participate in underlying cooperatively formed substructures. At such positions, even the apparently thermodynamically unfavorable introduction of hydrophobic residues should be explored.

L13 ANSWER 10 OF 184 MEDLINE on STN DUPLICATE 5
AB **alpha-Amylases**, in particular, microbial **alpha-amylases**, are widely used in industrial processes such as starch liquefaction and pulp processes, and more recently in detergency. Due to the need for **alpha-amylases** with high **specific activity** and activity at alkaline pH, which are critical parameters, for example, for the use in detergents, we have enhanced the **alpha-amylase** from **Bacillus** amyloliquefaciens (BAA). The genes coding for the wild-type BAA and the **mutants** BAA S201N and BAA N297D were subjected to error-prone PCR and gene shuffling. For the screening of **mutants** we developed a novel, reliable assay suitable for high throughput screening based on the Phadebas assay. One **mutant** (BAA 42) has an optimal activity at pH 7, corresponding to a shift of one pH unit compared to the wild type. BAA 42 is active over a broader pH range than the wild type, resulting in a 5-fold higher activity at pH 10. In addition, the activity in periplasmic extracts and the **specific activity**

increased 4- and 1.5-fold, respectively. Another **mutant** (BAA 29) possesses a wild-type-like pH profile but possesses a 40-fold higher activity in periplasmic extracts and a 9-fold higher **specific activity.** The comparison of the amino acid sequences of these two **mutants** with other homologous microbial **alpha-amylases** revealed the mutation of the highly conserved residues W194R, S197P, and A230V. In addition, three further mutations were found K406R, N414S, and E356D, the latter being present in other bacterial **alpha-amylases.**

L13 ANSWER 13 OF 184 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED. on STN DUPLICATE 8

AB The α -**amylase** from **Bacillus** licheniformis is the most widely used enzyme in the starch industry owing to its hyperthermostability, converting starch to medium-sized oligosaccharides. Based on sequence alignment of homologous amylases, we found a semi-conserved sequence pattern near the active site between transglycosidic and hydrolytic amylases, which suggested that hydrophobicity may play a role in modifying the transglycosylation/hydrolysis ratio. Based on this analysis, we replaced residue Val286 by Phe and Tyr in **Bacillus** licheniformis (x-amylase. Surprisingly, the two resultant **mutant** enzymes, Val286Phe and Val286Tyr, showed two different behaviors. Val286Tyr **mutant** was 5-fold more active for hydrolysis of starch than the wild-type enzyme. In contrast, the Val286Phe **mutant**, differing only by one hydroxyl group, was 3-fold less hydrolytic than the wild-type enzyme and apparently had a higher transglycosylation/hydrolysis ratio. These results are discussed in terms of affinity of subsites, hydrophobicity and electrostatic environment in the active site. The engineered enzyme reported here may represent an attractive alternative for the starch transformation industries as it affords direct and substantial material savings and requires no process modifications.

L13 ANSWER 14 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 9

AB **alpha-Amylases** (alpha-1,4-glucan-4-glucanohydrolases; EC 3.2.1.1) are classical **calcium**-containing enzymes, which constitute a family of endo-amylases catalysing the cleavage of alpha-D-(1-4) glycosidic bonds in starch and related carbohydrates with retention of the alpha-anomeric configuration in the products. They can be found in microorganisms, plants and higher organisms where they play a dominant role in carbohydrate metabolism. This study characterizes the substrate binding sites of **Bacillus** licheniformis **alpha-amylase** (BLA), human salivary **alpha-amylase** (HSA) and its Y151M **mutant.** It describes the first subsite maps, namely, number of subsites, position of cleavage sites and apparent subsite energies. The product pattern and cleavage frequencies were determined by HPLC, utilising a homologous series of chromophore-substituted maltooligosaccharides of degree of polymerisation (DP) 3 - 10 as model substrates. 2-Chloro-4-nitrophenyl (CNP) and 4,6-O-benzylidene-modified 4-nitrophenyl (Bnl-NP) beta-maltooligosaccharides (DP 4-8) were synthesised from cyclodextrins using a chemical procedure. For the preparation of CNP-maltooligosides of longer chain length a new chemoenzymatic procedure was developed using rabbit skeletal muscle glycogen phosphorylase b. Our results confirmed the presence of eight binding sites in BLA, five glycone sites (-5, -4, -3, -2, - 1), three aglycone sites (+1, +2, +3) and the catalytic site is located between subsites (- 1 and + 1). In addition, the subsite map revealed a barrier site at the reducing end of active site which repulses the glucose residue. The binding region of HSA is composed of four glycone and three aglycone-binding sites, while that of Tyr151Met **mutant** is composed of four glycone and two aglycone-binding sites. The subsite maps show that Y151M has strikingly decreased binding energy at subsite (+ 2), where the mutation has occurred (- 2.6 kJ/mol), compared to the binding energy at subsite (+ 2) of HSA (- 12.0 kJ/mol). (C) 2003 Elsevier

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L13 ANSWER 16 OF 184 MEDLINE on STN DUPLICATE 10
AB **Bacillus** licheniformis **alpha-amylase** (BLA) is a highly thermostable starch-degrading enzyme that has been extensively studied in both academic and industrial laboratories. For over a decade, we have investigated BLA thermal properties and identified amino acid substitutions that significantly increase or decrease the **thermostability.** This paper describes the cumulative effect of some of the most beneficial point mutations identified in BLA. Remarkably, the Q264S-N265Y double mutation led to a rather limited gain in **stability** but significantly improved the amylolytic function. The most hyperthermostable **variants** combined seven amino acid substitutions and inactivated over 100 times more slowly and at temperatures up to 23 degrees C higher than the wild-type enzyme. In addition, two highly destabilizing mutations were introduced in the metal binding site and resulted in a decrease of 25 degrees C in the half-inactivation temperature of the double **mutant** enzyme compared with wild-type. These mutational effects were analysed by protein modelling based on the recently determined crystal structure of a hyperthermostable BLA **variant.** Our engineering work on BLA shows that the **thermostability** of an already naturally highly thermostable enzyme can be substantially improved and modulated over a temperature range of 50 degrees C through a few point mutations.

L13 ANSWER 20 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
AB We have developed large-scale production of alkaline cellulases, alkaline proteases, and alkaline **alpha-amylases**, and the enzymes have been incorporated into heavy-duty compact detergents and/or bleaches. The problem with traditional detergent enzymes is that they are seriously inactivated by chemical oxidants and chelating reagents, and these enzymes are thermally unstable, especially when they are used in automatic dishwashers. We have found an alkaline liquefying **alpha-amylase** AmyK (formerly designated LAMY) from alkaliphilic **Bacillus** sp. strain KSM-1378. AmyK is highly active at alkaline pH, compared with other industrial **alpha-amylases** reported so far, and resistant to various surfactants. However, AmyK is less thermostable than the **Bacillus** licheniformis **alpha-amylase** (BLA), therefore, improvement in the **thermostability** of AmyK is desirable for use at high temperatures under alkaline conditions in automatic dishwashers. Moreover, AmyK and other **Bacillus alpha-amylases** are inactivated by chemical oxidants. We tried to improve the oxidative **stability** of AmyK by replacing a Met residue with non-oxidizable amino acids as in the case of alkaline proteases that acquired oxidative **stability** by site-directed mutagenesis. In this article, we describe the properties and deduced amino acid sequence of AmyK, and improvement in **thermostability** and oxidative **stability** of the enzyme by site-directed mutagenesis.

L13 ANSWER 30 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
AB High throughput screening of microbial DNA libraries was used to identify **alpha-amylases** with phenotypic characteristics compatible with large scale corn wet milling process conditions. Single and multiorganism DNA libraries originating from various environments were targeted for activity and sequence-based screening approaches. After initial screening, 15 clones were designated as primary hits based upon activity at pH 4.5 or 95 degreesC without addition of endogenous Ca2+. After further characterization, three enzyme candidates were chosen each with an exceptional expression of one or more aspects of the necessary phenotype: temperature **stability**, pH optimum, lowered reliance on Ca2+ and/or enzyme rate. To combine the best aspects of the three phenotypes to optimize process compatibility, the natural gene homologues were used as a parental sequence set for gene

reassembly. Approximately 21,000 chimeric daughter sequences were generated and subsets screened using a process-specific, high throughput activity assay. Gene reassembly resulted in numerous improved **mutants** with combined optimal phenotypes of expression, temperature **stability**, and pH optimum. After biochemical and process-specific characterization of these gene products, one a-amylase with exceptional process compatibility and economics was identified. This paper describes the synergistic approach of combining environmental discovery and laboratory evolution for identification and optimization of industrially important biocatalysts.

L13 ANSWER 32 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
AB A highly potent strain of **Bacillus** licheniformis 103 that synthesized thermostable α -**amylase** with temperature and pH optima of 90-95°C and 6.0-8.5, resp., was obtained by mutagenesis and selection. The composition of fermentation media and conditions for submerged cultivation of the producer were optimized. α -**Amylase** whose activity reached 260 U/mL was obtained in laboratory fermentors.

L13 ANSWER 34 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
AB A novel α -**amylase** (AmyK38) from an alkaliphilic **Bacillus** designated KSM-K38 is strongly resistant to chelators and oxidative reagents and contains no **calcium.** However, thermostabilization of AmyK38 is essential if it is to have industrial applications. Several chimeric enzymes between AmyK38 and the thermostable Arg181-Gly182-deleted **mutant** (dRG) of an . **alpha.-amylase** AmyK were constructed. A chimeric enzyme containing the N-terminal 21 amino acid residues of dRG was found to have higher **thermostability** than the parental AmyK38. By site-directed mutagenesis, AmyK38 was successfully thermostabilized by the single substitution of Tyr11 by Phe without any changes in the kinetic features.

L13 ANSWER 35 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
AB **Thermostability** and chelator resistance of the liquefying alkaline . **alpha.-amylase** (AmyK) from alkaliphilic **Bacillus** sp. strain KSM-1378 were examined by deletion of either Arg181-Gly182 or Thr 183-Gly184 on a loop in domain B. In the tertiary structure of **Bacillus** stearothermophilus α -**amylase** (BSA), Ile181-Gly182 (Thr183-Gly184 in AmyK) pushes away a spatially contacting region containing Ca2+-coordinating Asp207 (Asp209 in AmyK). Therefore, the deletion of Ile181-Gly182 rather than Arg179-Gly180 was predicted to result in a higher **thermostability** of BSA. However, our results with AmyK were clearly contrary to this prediction. The resistance to EDTA of both **mutant** enzymes from AmyK was essentially equal, and the Arg181-Gly182-deleted **mutant** was more thermostable than the Thr183-Gly184-deleted one. It strongly implies that the microenvironmental topol. around the loop containing these dipeptides in AmyK is different from that in BSA.

L13 ANSWER 36 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 18
AB **Bacillus** licheniformis a-amylase (BLA) is a highly thermostable enzyme which is widely used in biotechnological processes. Although it is produced by a non-thermophilic bacterium, it remains active for several hours at temperatures over 90degreesC under conditions of industrial starch hydrolysis. It is also far more thermostable than the **alpha-amylases** from B. stearothermophilus and B. amyloliquefaciens despite the strong sequence similarities between these three proteins. BLA provides therefore an interesting model for protein engineers investigating enzyme **thermostability** and thermostabilization. Over the last decade, we have performed an extensive

mutational and structural analysis on BLA in order to elucidate the origin of its unusual thermal properties and, if possible, increase its **thermostability** even further. Before the three-dimensional structure was known, we had used "blind" mutagenesis and identified two critical positions where amino-acid substitutions could either increase or decrease significantly the rate of irreversible thermoinactivation. Once a detailed X-ray structure of BLA was solved, structure-based mutagenesis was used to probe the role of residues involved in salt-bridges, **calcium**-binding or potential deamidation processes. Our results revealed the key role of domain B and its interface with domain A in determining the overall **thermostability** of BLA. Most of the mutations we introduced in this region modify the **stability** in one way or another by influencing the network of electrostatic interactions entrapping a Ca-Na-Ca metal triad at the domain A/B interface. In the course of this mutational study we have constructed over 500 BLA **variants** bearing single or multiple mutations, among which many were found to be either highly detrimental or slightly beneficial to the **stability.** The cumulative effect of the mutations enabled us to modulate the enzyme **stability** over a 50degreesC temperature range without perturbing significantly the amylolytic function. Although a full understanding of the origin of BLA natural thermoresistance has not yet been reached, our study demonstrated that it is not optimized and that it can be increased or decreased artificially by several means.

L13 ANSWER 37 OF 184 MEDLINE on STN DUPLICATE 19

AB The **alpha-amylase** from **Bacillus** sp. strain TS-23 is a secreted starch hydrolase with a domain organization similar to that of other microbial **alpha-amylases** and an additional functionally unknown domain (amino acids 517-613) in the C-terminal region. By sequence comparison, we found that this latter domain contained a sequence motif typical for raw-starch binding. To investigate the functional role of the C-terminal region of the **alpha-amylase** of **Bacillus** sp. strain TS-23, four His(6)-tagged **mutants** with extensive deletions in this region were constructed and expressed in Escherichia coli. SDS-PAGE and activity staining analyses showed that the N- and C-terminally truncated **alpha-amylases** had molecular masses of approximately 65, 58, 54, and 49 kDa. Progressive loss of raw-starch-binding activity occurred upon removal of C-terminal amino acid residues, indicating the requirement for the entire region in formation of a functional starch-binding domain. Up to 98 amino acids from the C-terminal end of the **alpha-amylase** could be deleted without significant effect on the raw-starch hydrolytic activity or thermal **stability** . Furthermore, the active **mutants** hydrolyzed raw corn starch to produce maltopentaose as the main product, suggesting that the raw-starch hydrolytic activity of the **Bacillus** sp. strain TS-23 **alpha-amylase** is functional and independent from the starch-binding domain.

L13 ANSWER 43 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN

AB The invention provides a method of obtaining improved proteins, particularly enzymes, having altered **stability** characteristics, especially thermal **stability.** Such protein is modified from a precursor amino acid sequence by the substitution or deletion of an amino acid residue which differs from a corresponding amino acid residue in a less stable but homologous protein, wherein said improved protein has improved properties compared to a protein corresponding to the precursor amino acid sequence.

L13 ANSWER 55 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN

AB Recombinant **mutant** α -**amylase** with improved **thermostability**, its recombinant expression, and detergent containing it, are disclosed. α -**Amylase**

(LAMY) from alkalophilic **Bacillus** sp. strain KSM-1378 is a novel semi-alkaline enzyme which has 5-fold higher **specific activity** than that of a **Bacillus** licheniformis enzyme. The Ile193 in LAMY was replaced with aspartic acid (I193D) by site-directed mutagenesis to increase **thermostability** of the enzyme. **Thermostability** was further increased by deletion of Arg181 and Gly182 (RG+I193D). Dishwasher detergent containing I193D or RG+I193D showed superior cleansing ability.

L13 ANSWER 56 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
AB α **-Amylase mutants** having lowered **thermostability** as compared to their wild type are prepared from the α **-amylase** of **Bacillus** amyloliquefaciens clone 21 to suit for the preparation of bakery products. The **mutants** remain <80% active after incubating at 65° for 30 min. The **mutants** are prepared by substitution mutations at 380-Ala→Thr and 393-Phe→ser; 30-Ser→Leu and 195-Asp→Asn; 154-Arg→Lys; and 192-Ala→Val and 233-Asp→Asn; resp. The **mutants** improve the bread quality.

L13 ANSWER 59 OF 184 MEDLINE on STN DUPLICATE 28
AB **Bacillus** licheniformis **alpha-amylase** (BLA) is a starch-degrading enzyme that is highly thermostable although it is produced by a rather mesophilic organism. Over the last decade, the origin of BLA thermal properties has been extensively investigated in both academic and industrial laboratories, yet it is poorly understood. Here, we have used structure-based mutagenesis in order to probe the role of amino acid residues previously proposed as being important for BLA **thermostability.** Residues involved in salt-bridges, **calcium** binding or potential deamidation processes have been selected and replaced with various amino acids using a site-directed mutagenesis method, based on informational suppression. A total of 175 amylase **variants** were created and analysed in vitro. Active amylase **variants** were tested for **thermostability** by measuring residual activities after incubation at high temperature. Out of the 15 target residues, seven (Asp121, Asn126, Asp164, Asn192, Asp200, Asp204 and Ala269) were found to be particularly intolerant to any amino acid substitutions, some of which lead to very unstable **mutant** enzymes. By contrast, three asparagine residues (Asn172, Asn188 and Asn190) could be replaced with amino acid residues that significantly increase the **thermostability** compared to the wild-type enzyme. The highest stabilization event resulted from the substitution of phenylalanine in place of asparagine at position 190, leading to a sixfold increase of the enzyme's half-life at 80 degrees C (pH 5.6, 0.1 mM CaCl(2)).These results, combined with those of previous mutational analyses, show that the structural determinants contributing to the overall **thermostability** of BLA concentrate in domain B and at its interface with the central A domain. This region contains a triadic Ca-Na-Ca metal-binding site that appears extremely sensitive to any modification that may alter or reinforce the network of electrostatic interactions entrapping the metal ions. In particular, a loop spanning from residue 178 to 199, which undergoes pronounced conformational changes upon removal of **calcium**, appears to be the key feature for maintaining the enzyme structural integrity. Outside this region, most salt-bridges that were destroyed by mutations were found to be dispensable, except for an Asp121-Arg127 salt-bridge that contributes to the enhanced **thermostability** of BLA compared to other homologous bacterial **alpha-amylases.** Finally, our studies demonstrate that the natural resistance of BLA against high temperature is not optimized and can be enhanced further through various means, including the removal of possibly deamidating residues.
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L13 ANSWER 60 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
AB **Calcium** independent and acid stable **alpha** -**amylases** for starch liquefaction were developed by protein engineering. Termamyl LC(TM) obtained by site-directed mutagenesis showed high **calcium** independence, and its performance in the absence of **calcium** is equal to the one with Termamyl(TM) in the presence of 40 ppm of **calcium.** Termamyl LC(TM) was further developed by random mutagenesis, and highly improved **variants** have been efficiently produced by recent protein engineering technologies.

The development of detergent **alpha** -**amylase** began with microbial screening, and two **alpha** -**amylases** active and stable in alkaline conditions were identified. Those amylases were further developed by protein engineering (site-directed mutagenesis), resulting in **variants** with improved alkaline **stability** and **calcium** independence.

L13 ANSWER 75 OF 184 MEDLINE on STN DUPLICATE 33
AB Industrial-scale starch liquefaction is currently constrained to operating at pH 6.0 and above, as the enzyme used in the process, **Bacillus** licheniformis **alpha-amylase**, is unstable at lower pH under the conditions used. There is a need to develop an enzyme that can operate at lower pH. Recent progress has been made in engineering the B. licheniformis enzyme for improved industrial performance. The availability of crystal structures and subsequent analysis of improved **variants**, in a structural context, is revealing common factors and a rationale to make further improvements.

L13 ANSWER 77 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
AB Use of specific **alpha-amylase** (EC-3.2.1.1) enzymes in a laundry surfactant composition is claimed, where the **alpha-amylase** has: a **specific activity** of at least 25% higher than the **specific activity** of Termamyl at a temperature range of 25-55 deg and at pH 8-10, measured by the Phadebas **alpha-amylase** assay; a disclosed protein sequence or has at least 80% homology to the protein sequence; the following protein sequence in the N-terminus His-His-Asn- Gly-Thr-Asn- Gly-Thr-Met-Met-Gln-Tyr- Phe-Glu-Trp- Tyr-Leu-Pro- Asn-Asp or at least 80% homology to this sequence; been derived from an alkalophilic **Bacillus** sp., especially strains NCIB 12289, NCIB 12512, NCIB 12513, and DSM 935; immunological crossreactivity with antibodies against the disclosed protein sequence; or a **variant** (deletion, insertion or substitution **mutant**) compared to a parent **alpha-amylase** (where the **variant** is encoded by a disclosed DNA sequence, which hybridizes to a DNA probe). The **variant** has increased **thermostability**, increased **stability** toward oxidation, reduced Ca ion dependency, increased **stability** and/or increased **alpha-amylase** activity at neutral to relatively high pH. (81pp)

L13 ANSWER 89 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
AB Many recent approaches involving site-directed **mutants** have succeeded in increasing the **thermostability** of proteins. It is well known that replacements with proline residues reduce the conformational degrees of freedom in the main polypeptide chain and thus can increase protein thermostabilization. We have studied protein thermostabilization by introducing proline substitutions in the homologous oligo-1,6-glucosidases from various **Bacillus** strains which grow within different temperature ranges. As a consequence, the 'proline rule' was proposed for protein thermostabilization. The principle of this rule is that an increase in the frequency of proline occurrence at beta-turns and/or an increase in the total number of hydrophobic residues can enhance protein **thermostability.** We have generated several lines of evidence supporting the theory from the comparative analysis of oligo-1,6-glucosidases in their primary and secondary structures and

molecular properties, the X-ray crystal structure analysis of the **Bacillus** cereus oligo-1,6-glucosidase, and the enhancement in **thermostability** of the oligo-1,6-glucosidase by cumulative replacements with prolines. As a new finding from the studies, two specific sites (second positions at beta-turns and N1 positions of alpha-helices) were found to be the most critical to protein thermostabilization dependent on several structural prerequisites for proline substitution. (C) 1998 Elsevier Science B.V. All rights reserved.

L13 ANSWER 94 OF 184 MEDLINE on STN DUPLICATE 43
AB This paper provides further understanding of the thermodynamic and structural features determining the **stability** of **Bacillus** licheniformis **alpha-amylase** (BLA) at two crucial positions, His133 and Ala209. Results of protein modelling and saturated site-directed mutagenesis at position 133 and 209 have been reported in a previous paper (Declerck et al., 1995, Prot. Engng, 8, 1029-1037). In the first part of the present work, evidence is presented supporting the hypothesis that the stabilizing mutations reduce the rate of initial unfolding of the enzyme during the reversible step of the inactivation reaction and do not modify the irreversible processes undergone subsequently by the unfolded molecules. In the second part, we have examined the three-dimensional structure of BLA which has been determined recently by X-ray analysis (Machius et al., 1995, J. Mol. Biol., 246, 545-559). This analysis showed that our previous predictions made from molecular modelling were partly correct. At position 209, the effect of the stabilizing substitutions can be explained by a groove-filling effect reinforcing the hydrophobic packing between two helices of the central domain, while preserving a well-ordered water structure at the surface. At position 133, the stabilizing substitutions must compensate the loss of the hydrogen bond network in which the original histidine side-chain is involved; this compensation could be achieved through enhanced hydrophobic side-chain interactions within the beta-sheet where residue 133 is located, which correlates with the propensity of the residue to form and maintain a beta-strand conformation of the main chain at this position.

L13 ANSWER 95 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
AB An improved strain was developed for hyperproduction of a thermostable **alpha-amylase** (AA, EC-3.2.1.1). **Bacillus** stearothermophilus MK716 grew optimally at 55 deg (maximum 70 deg) and pH 7.0 (range 5-8). AA production was induced by starch and repressed by glucose, and was growth-related. The AA was optimally active at pH 5.6 and 70 deg, with 15% activity at 100 deg and 80% at pH 6.4. The AA had higher activity at 70 deg than Ban AA, and equivalent activity at 83 deg to Termamyl AA. The enzyme also had greater activity at pH 5.6-6.4. When the strain was subjected to ethylmethane sulfonate mutagenesis, **mutant** E1 was obtained, which produced 40-fold more AA. Through cloning and subcloning in Escherichia coli TB1, a 2.0 kb fragment was found to be sufficient for expression and secretion of AA, and was in **Bacillus** subtilis. Subclone BGAT9, containing recombinant plasmid pAmyB9 with a 2.0 kb insert, produced 107 times more AA than MK716. pAmyB9 was completely stable in B. subtilis. Cloning in B. subtilis did not affect **thermostability** of the AA, but widened its optimum activity range to pH 5.6-6.4. (34 ref)

L13 ANSWER 104 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN
AB The thermal **stability** of proteins was studied, 195 single amino acid residue replacements reported elsewhere being analysed for several protein conformational characteristics: type of residue replacement; conservative versus nonconservative substitution; replacement being in a homologous stretch of amino acid residues; change in hydrogen bond, van der Waals and secondary structure propensities; solvent-accessible versus inaccessible replacement; type of secondary structure involved in the substitution; the physico-chemical

characteristics to which the **thermostability** enhancement can be attributed; and the relationship of the replacement site to the folding intermediates of the protein, when known. From the above analyses, some general rules arise which suggest where amino acid substitutions can be made to enhance protein **thermostability**: substitutions are conservative according to the Dayhoff matrix; mainly occur on conserved stretches of residues; preferentially occur on solvent-accessible residues; maintain or enhance the secondary structure propensity upon substitution; contribute to neutralize the dipole moment of the caps of helices and strands; and tend to increase the number of potential hydrogen bonding or van der Waals contacts or improve hydrophobic packing.

L13 ANSWER 105 OF 184 HCAPLUS COPYRIGHT 2004 ACS on STN
AB B. licheniformis α -**amylase** (I) is a major industrial enzyme used for the hydrolysis of starch at high temperature By genetic engineering, hyperthermostable **mutants** of this highly thermostable enzyme could be obtained, bearing mutations at 2 crucial positions, His-133 and Ala-209. The results of protein modeling and saturated directed mutagenesis at these 2 sites were reported in a recent paper from the authors' laboratory The present work provides further understandings of the thermodn. and structural features determining the **stability** of I at positions His-133 and Ala-209. Evidences are presented supporting the hypothesis that the stabilizing mutations reduce the rate of initial unfolding of the enzyme during the reversible step of the inactivation reaction and do not modify the irreversible processes undergone subsequently by the unfolded mols. The authors examined the 3-dimensional model of I which was recently determined by x-ray anal. This showed that their previous predictions made from mol. modeling were mostly correct. At position 133, the stabilizing substitutions must compensate the loss of the H-bond network in which the original His side-chain is implicated. This could be related to the propensity of the inserted amino acid at forming β-sheet and increasing hydrophobic interactions within the β-sheet region where residue 133 is located. At position 209, the effect of the stabilizing substitutions could be mostly explained by a groove-filling effect reinforcing the hydrophobic packing between 2 helixes of the central domain while preserving a well-ordered water structure a the surface.

L13 ANSWER 108 OF 184 MEDLINE on STN DUPLICATE 50
AB We have identified previously two critical positions for the **thermostability** of the highly thermostable **alpha-amylase** from **Bacillus** licheniformis. We have now introduced all 19 possible amino acid residues to these two positions, His133 and Ala209. The most favourable substitutions were to Ile and Val, respectively, which both increased the half-life of the enzyme at 80 degrees C by a factor of approximately 3. At both positions a stabilizing effect of hydrophobic residues was observed, although only in the case of position 133 could a clear correlation be drawn between the hydrophobicity of the inserted amino acid and the gain in protein **stability.** The construction of double **mutants** showed a cumulative effect of the most favourable and/or deleterious substitutions. Computer modelling was used to generate a 3-D structure of the wild-type protein and to model substitutions at position 209, which lies in the conserved (alpha/beta)8 barrel domain of **alpha-amylase**; Ala209 would be located at the beginning of the third helix of the barrel, in the bottom of a small cavity facing the fourth helix. The model suggests that replacement by, for example, a valine could fill this cavity and therefore increase intra- and interhelical compactness and hydrophobic interactions.

L13 ANSWER 110 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 51
AB An **alpha-amylase** from a hyper-producing strain of **Bacillus** (sp. E2) was stable at 70 degrees C for 30 min but was

quickly inactivated at higher temperatures. In the presence of 10 mM Ca2+ and starch (20% w/v), however, the enzyme was stable at 90 degrees C for 10 min and after 30 min at 100 degrees C still retained 26% of its initial activity.

L13 ANSWER 119 OF 184 MEDLINE on STN DUPLICATE 56
AB A series of truncated proteins from a thermostable **Bacillus** stearothermophilus **alpha-amylase** was prepared to study the importance of the extension in the C-terminus compared with other liquefying **Bacillus alpha-amylases.** The mutations introducing new translation termination sites shortened the 515 amino acid residue-long wild type enzyme by 17, 32, 47, 73 or 93 residues. The longer the truncation, the lower the **specific activity** of the enzyme. Only the two longest **mutant** proteins were active: the **specific activity** of the 498 residue **variant** was 97% and protein 483 was 36% that of the parental enzyme. The Km values of starch hydrolysis changed from 1.09 for wild type enzyme to 0.35 and 0.21 for **mutants** 498 and 483, respectively, indicating altered substrate binding. The **mutant** enzymes had almost identical pH and temperature optima with the wild type amylase, but enhanced thermal **stability** and altered end product profile. The consequences of the truncation to the structure and function of the enzymes were explored with molecular modeling. The liquefying amylases seem to require approximately 480 residues to be active, whereas the C-terminal end of B.stearothermophilus amylase is required for increased activity.

L13 ANSWER 121 OF 184 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN DUPLICATE 57
AB The **alpha-amylase** gene of **Bacillus** licheniformis has been cloned and two **mutant alpha-amylase** genes of which histidine 235 was changed to glutamine (H235Q) and aspartic acid 328 to glutamic acid (D328E) have been produced by site-directed mutagenesis. The kinetic parameters, optimum pH and **thermostability** of wild type(WT) and these two **mutant** amylases expressed in E. coli MC1061 have been compared after purification. The K-m values of WT, H235Q and D328E **alpha-amylases** were 0.22%, 0.73%, and 0.80%, respectively, when using starch as the substrate. The V-max values of wild type **alpha-amylase** and **mutant alpha-amylases** were 0.6-0.7%/minute, and did not show any significant differences among them. The optimum pH of D328E **alpha-amylase** was shifted to more acidic pH. Also, the **thermostability** of H235Q **alpha-amylase** was increased compared to the wild type **alpha-amylase.**

L13 ANSWER 126 OF 184 LIFESCI COPYRIGHT 2004 CSA on STN DUPLICATE 58
AB Crude **alpha -amylase** was obtained from culture supernatant of **Bacillus** licheniformis **mutant** 7902. Enzyme activity increased as the temperature raised gradually from 75 to 100 degree C. The enzyme was fairly stable retaining more than 90% of its original activity after 60 min at 90 degree C and 20 min at 95 degree C. The enzyme was purified by ammonium sulfate fractionation, Sephadex G-50 gel filtration and polyacrylamide slab gel electrophoresis. The **specific activity** of purified enzyme was 49.3 fold of the crude enzyme. The purified **alpha -amylase** was identified to be homogeneous by SDS electrophoresis. Molecular weight of this enzyme was 68000. Ca super(2+), Li super(+) and Mg super(2+) ions enhanced the enzyme activity, whereas Al super(3+), Ag super(+), Cu super(2+) and Fe super(2+) inhibited it.

L13 ANSWER 131 OF 184 MEDLINE on STN DUPLICATE 62
AB Site-directed mutagenesis of **Bacillus** subtilis N7 **alpha -amylase** has been performed to evaluate the roles of the active

site residues in catalysis and to prepare an inactive catalytic-site **mutant** that can form a stable complex with natural substrates. Mutation of Asp-176, Glu-208, and Asp-269 to their amide forms resulted in over a 15,000-fold reduction of its **specific activity**, but all the **mutants** retained considerable substrate-binding abilities as estimated by gel electrophoresis in the presence of soluble starch. Conversion of His-180 to Asn resulted in a 20-fold reduction of kcat with a 5-fold increase in Km for a maltopentaose derivative. The relative affinities for acarbose vs. maltopentaose were also compared between the **mutants** and wild-type enzyme. The results are consistent with the roles previously proposed in Taka-amylase A and porcine pancreatic **alpha-amylase** based on their X-ray crystallographic analyses, although different pairs had been assigned as catalytic residues for each enzyme. Analysis of the residual activity of the catalytic-site **mutants** by gel electrophoresis has suggested that it derived from the wild-type enzyme contaminating the **mutant** preparations, which could be removed by use of an acarbose affinity column; thus, these **mutants** are completely devoid of activity. The affinity-purified **mutant** proteins should be useful for elucidating the complete picture of the interaction of this enzyme with starch.

L13 ANSWER 137 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
AB A **mutant** enzyme (I) is claimed which is produced by microbial fermentation and exhibits reduced **stability** under industrial conditions relative to the wild-type enzyme. (I) is a bacterial **alpha-amylase** (AA, EC-3.2.1.1) obtained by at least 1 selected mutation of wild-type AA, and exhibits bread improving properties and reduced **thermostability** during baking. (I) comprises a protein sequence differing by 1-10 amino acids from that of the wild-type AA, preferably with Arg123 replaced by Cys. Alternatively, (I) is **Bacillus** amyloliquefaciens AA with a mutation at at least 1 of amino acids 113, 114, 116, 123, 163, 164, 166, 238, 316, 322, 345, 349, 356, 386, 394 or 398. Modified B. amyloliquefaciens AA, dough or similar products and bread or related products produced using (I), microorganisms which have been made suitable for (I) production by elimination or inactivation of endogenous AA or transformation with a gene encoding (I), a gene encoding (I), a vector plasmid containing the gene, and a bread improver composition, are also claimed. (I) is cheap to produce and improves bread crumb softness and loaf volume without starch dextrinization. (26pp)

L13 ANSWER 141 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
AB A new **alpha-amylase** (EC-3.2.1.1) was isolated from a **Bacillus** licheniformis **mutant.** The enzyme was capable of catalyzing industrial scale starch liquefaction at lower than conventional pH levels (optimum 5.5-6), resulting in significant cost savings and less complex operations. Liquefaction studies in a pilot plant jet cooker showed that commercial starch slurries taken from different sources varied greatly in ease of liquefaction at lower than conventional pH values. Low levels of stabilizing or destabilizing factors appeared to exist in commercial starch slurries, which affected the **stability** of **alpha-amylase** during high-temperature (103-107 deg) liquefaction. The new starch liquefaction process avoided the formation of maltulose during liquefaction, and ionexchange requirements were decreased. However, further work is required before liquefaction may be carried out under saccharification conditions, or in the absence of **calcium** addition. (0 ref)

L13 ANSWER 146 OF 184 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
AB The relationship between structure, activity and **stability** of thermostable **Bacillus** stearothermophilus **alpha-amylase** (EC-3.2.1.1) was studied by site-directed mutagenesis. The functions of the conserved amino acids were examined by replacing

Arg232, His328 and Asp331 with Lys232, Asp238 and Glu331, respectively. The mutated proteins were expressed in **Bacillus** subtilis, purified and characterized. Mutation of His328, involved in **calcium**- and substrate-binding, to Asp238 reduced the **specific activity** by 47% and lowered the inactivation temperature remarkably. The end-product profile of the **mutant** enzyme was shifted towards shorter end-products e.g. glucose and maltose. Replacing the active site Asp331 with Glu331 resulted in almost complete inactivation of the enzyme. This **mutant** liberated maltose and maltotriose from starch after prolonged incubation. Replacing Arg232 with Lys232 lowered the **specific activity** by about 80%. The **mutant** enzyme exhibited almost the same **thermostability** as the wild-type enzyme, but had a much broader pH optimum profile (pH 4.5-7.0) compared to the wild-type (pH 4.5-5.5). (0 ref)

L13 ANSWER 149 OF 184 MEDLINE on STN DUPLICATE 69
AB The relationship between structure, activity, and **stability** of the thermostable **Bacillus** stearothermophilus **alpha-amylase** was studied by site-directed mutagenesis of the three most conserved residues. Mutation of His-238 to Asp involved in Ca2+ and substrate binding reduced the **specific activity** and thermal **stability**, but did not affect the pH and temperature optima. Replacement of Asp-331 by Glu in the active site caused almost total inactivation. Interestingly, in prolonged incubation this **mutant** enzyme showed an altered end-product profile by liberating only maltose and maltotriose. Conservative mutation of the conserved Arg-232 by Lys, for which no function has yet been proposed, resulted in lowered **specific activity**: around 12% of the parental enzyme. This **mutant** enzyme had a wider pH range but about the same temperature optimum and thermal **stability** as the wild-type enzyme. Results obtained with different **mutants** were interpreted by computer aided molecular modeling.

L13 ANSWER 155 OF 184 MEDLINE on STN DUPLICATE 73
AB The oligonucleotide encoding Bam HI recognition site having the structure pCGGGATC had been inserted into the recognition sites MspI of the B. amyloliquefaciens **alpha-amylase** gene, which was cloned in pTG29B plasmid. The **alpha-amylase** gene had no BamHI sites before mutagenesis. The set of pNSBamHI plasmids with BamHI site at four different positions was obtained. It was shown that all the **mutant alpha-amylases** possess different specific activities. One of the **mutant** proteins possesses reduced **thermostability.** The **mutant alpha-amylases** can be used for further experiments on protein-engineering of liquefying-type **alpha-amylases.**

L13 ANSWER 156 OF 184 MEDLINE on STN DUPLICATE 74
AB **Alpha-amylase** genes of **Bacillus** amyloliquefaciens, coding proteins with reduced **thermostability**, had been obtained as a result of hydroxylamine mutagenesis. Temperature, pH and starch concentration dependences of two **mutant alpha-amylases** were investigated. The synthesis of the **alpha-amylases** by several B. subtilis strains with different levels of extracellular proteases was also studied. The mutation containing fragments were localized and the structures of the mutations were determined. It was found that the decrease of **thermostability** of **mutant** No 141 was due to Asp to Asn change at the position No 194 of the mature protein, and for **mutant** No 191--due to Glu to Lys change at the position No 185.

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S18	632	(alpha adj amylase$1) same bacillus same (mutant$1 or variant$1)	US-PGPUB; USPAT	OR	OFF	2004/05/07 13:41
S19	11197	(mutant$1 or variant$1) same (stability or thermostabilty or specific adj activity or calcium)	US-PGPUB; USPAT	OR	OFF	2004/05/07 13:42
S20	332	S18 and S19	US-PGPUB; USPAT	OR	OFF	2004/05/07 13:44